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(71) Applicant: NEXSTAR PHARMACEUTICALS, INC. [US/US]; Suite 200, 2860 Wilderness Place, Boulder, CO 80301 (US).

(72) Inventors: GOLD, Larry; 1033 5th Street, Boulder, CO 80302 (US). PAGRATIS, Nikos; 5813 North Orchard Creek Circle, Boulder, CO 80301 (US).

(74) Agents: SWANSON, Barry, J. et al.; Swanson & Bratschun, L.L.C., Suite 200, 8400 E. Prentice Avenue, Englewood, CO 80111 (US).

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(54) Title: HIGH AFFINITY TGFβ NUCLEIC ACID LIGANDS AND INHIBITORS

(57) Abstract

Methods are described for the identification and preparation of high–affinity nucleic acid ligands to TGFβ. Included in the invention are specific RNA ligands to TGFβ1 identified by the SELEX method. Also included are RNA ligands that inhibit the interaction of TGFβ1 with its receptor.

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HIGH AFFINITY TGFß NUCLEIC ACID LIGANDS AND INHIBITORS

FIELD OF THE INVENTION

Described herein are methods for identifying and preparing high-affinity nucleic acid ligands to TGFß. The method utilized herein for identifying such nucleic acid ligands is called SELEX, an acronym for Systematic Evolution of Ligands by EXponential enrichment. This invention includes high affinity nucleic acid ligands of TGFß. Further disclosed are RNA ligands to TGFß1. Also included are oligonucleotides containing nucleotide derivatives chemically modified at the 2'-positions of pyrimidines. Additionally disclosed are RNA ligands to TGFß1 containing 2'-F-modifications. This invention also includes high affinity nucleic acid inhibitors of TGFß1. The oligonucleotides of the present invention are useful as pharmaceuticals or diagnostic agents.

BACKGROUND OF THE INVENTION

The transforming growth factor -ß (TGFß) polypeptides influence growth, differentiation, and gene expression in many cell types. The first polypeptide of this family that was characterized, TGFß1 has two identical 112 amino acid subunits which are covalently linked. TGFß1 is a highly conserved protein with only a single amino acid difference distinguishing humans from mice. There are two other members of the TGFß gene family that are expressed in mammals. TGFß2 is 71% homologous to TGFß1 (de Martin *et al.* (1987) EMBO J. 6:3673-3677), whereas TGFß3 is 80% homologous to TGFß1(Derynck *et al.* (1988) EMBO J 7:3737-3743). The structural characteristics of TGFß1 as determined by nuclear magnetic resonance (Archer *et al.* (1993) Biochemistry 32:1164-1171) agree with the crystal structure of TGFß2 (Daopin *et al.* (1992) Science 257:369-374; Schlunegger and Grutter (1992) Nature 358:430-434).

Even though the TGFß's have similar three dimensional structures, they are by no means physiologically equivalent. There are at least three different extracellular receptors, type I, II and III, involved in transmembrane signaling of TGFß to cells carrying the receptors. (For reviews, see Derynck (1994) TIBS 19:548-553 and Massague (1990) Ann.

Rev. Cell Biol. 6:597-641). In order for TGFß2 to effectively interact with the type II TGFß receptor, the type III receptor must also be present (Derynck (1994) TIBS 19:548-553). Vascular endothelial cells lack the type III receptor. Instead endothelial cells express a structurally related protein called endoglin (Cheifetz *et al.* (1992) J. Biol. Chem. 267:19027-19030), which only binds TGFß1 and TGFß3 with high affinity. Thus, the relative potency of the TGFß's reflect the type of receptor expressed in a cell and organ system.

In addition to the regulation of the components in the multifactorial signaling pathway, the distribution of the synthesis of TGFß polypeptides also affects physiological function. The distribution of TGFß2 and TGFß3 is more limited (Derynck *et al.* (1988) EMBO J 7:3737-3743) than TGFß1, e.g., TGFß3 is limited to tissues of mesenchymal origin, whereas TGFß1 is present in both tissues of mesenchymal and epithelial origin.

TGFß1 is a multifunctional cytokine critical for tissue repair. High concentrations of TGFß1 are delivered to the site of injury by platelet granules (Assoian and Sporn (1986) J. Cell Biol. 102:1217-1223). TGFß1 initiates a series of events that promote healing including chemotaxis of cells such as leukocytes, monocytes and fibroblasts, and regulation of growth factors and cytokines involved in angiogenesis, cell division associated with tissue repair and inflammatory responses. TGFß1 also stimulates the synthesis of extracellular matrix components (Roberts *et al.* (1986) Proc. Natl. Acad. Sci. USA 83:4167-4171; Sporn *et al.* (1983) Science 219:1329-1330; Massague (1987) Cell 49:437-438) and most importantly for understanding the pathophysiology of TGFß1, TGFß1 autoregulates its own synthesis (Kim *et al.* (1989) J. Biol. Chem. 264:7041-7045).

A number of diseases have been associated with TGFß1 overproduction. Fibrotic diseases associated with TGFß1 overproduction can be divided into chronic conditions such as fibrosis of the kidney, lung and liver and more acute conditions such as dermal scarring and restenosis. Synthesis and secretion of TGFß1 by tumor cells can also lead to immune suppression such as seen in patients with aggressive brain or breast tumors (Arteaga *et al.* (1993) J. Clin. Invest. 92:2569-2576). The course of Leishmanial infection in mice is drastically altered by TGFß1 (Barral-Netto *et al.* (1992) Science 257:545-547). TGFß1 exacerbated the disease, whereas TGFß1 antibodies halted the progression of the disease in genetically susceptible mice. Genetically resistant mice became susceptible to Leishmanial infection upon administration of TGFß1.

The profound effects of TGFß1 on extracellular matrix deposition have been reviewed (Rocco and Ziyadeh (1991) in Contemporary Issues in Nephrology v.23, "Hormones, Autocoids and the Kidney," ed. Jay Stein, Churchill Livingston, NY pp.391-410; Roberts *et al.* (1988) Rec. Prog. Hormone Res. 44:157-197) and include the stimulation of the synthesis and the inhibition of degradation of extracellular matrix components. Since the structure and filtration properties of the glomerulus are largely determined by the extracellular matrix composition of the mesangium and glomerular membrane, it is not surprising that TGFß1 has profound effects on the kidney. The accumulation of mesangial matrix in proliferative glomerulonephritis (Border *et al.* (1990) Kidney Int. 37:689-695) and diabetic nephropathy (Mauer *et al.* (1984) J. Clin. Invest. 74:1143-1155) are clear and dominant pathological features of the diseases. TGFß1 levels are elevated in human diabetic glomerulosclerosis (advanced neuropathy) (Yamamoto *et al.* (1993) Proc. Natl. Acad. Sci. USA 90:1814-1818). TGFß1 is an important mediator in the genesis of renal fibrosis in a number of animal models (Phan *et al.* (1990) Kidney Int. 37:426; Okuda *et al.* (1990) J. Clin. Invest. 86:453). Suppression of experimentally induced glomerulonephritis in rats has been demonstrated by antiserum against TGFß1 (Border *et al.* (1990) Nature 346:371) and by an extracellular matrix protein, decorin, which can bind TGFß1 (Border *et al.* (1992) Nature 360:361-363).

Too much TGFß1 leads to dermal scar-tissue formation. Neutralizing TGFß1 antibodies injected into the margins of healing wounds in rats have been shown to inhibit scarring without interfering with the rate of wound healing or the tensile strength of the wound (Shah *et al.* (1992) Lancet 339:213-214). At the same time there was reduced angiogenesis, reduced number of macrophages and monocytes in the wound, and a reduced amount of disorganized collagen fiber deposition in the scar tissue.

TGFß1 may be a factor in the progressive thickening of the arterial wall which results from the proliferation of smooth muscle cells and deposition of extracellular matrix in the artery after balloon angioplasty. The diameter of the restenosed artery may be reduced 90% by this thickening, and since most of the reduction in diameter is due to extracellular matrix rather than smooth muscle cell bodies, it may be possible to open these vessels to 50% simply by reducing extensive extracellular matrix deposition. In uninjured pig arteries transfected *in vivo* with a TGFß1 gene, TGFß1 gene expression was associated with both extracellular matrix synthesis and hyperplasia (Nabel *et al.* (1993) Proc. Natl.

Acad. Sci. USA 90:10759-10763). The TGFß1 induced hyperplasia was not as extensive as that induced with PDGF-BB, but the extracellular matrix was more extensive with TGFß1 transfectants. No extracellular matrix deposition was associated with FGF-1 (a secreted form of FGF) induced hyperplasia in this gene transfer pig model (Nabel (1993) Nature 362:844-846).

There are several types of cancer where TGFß1 produced by the tumor may be deleterious. MATLyLu rat cancer cells (Steiner and Barrack (1992) Mol. Endocrinol. 6:15-25) and MCF-7 human breast cancer cells (Arteaga *et al.* (1993) Cell Growth and Differ. 4:193-201) became more tumorigenic and metastatic after transfection with a vector expressing the mouse TGFß1. In breast cancer, poor prognosis is associated with elevated TGFß (Dickson *et al.* (1987) Proc. Natl. Acad. Sci. USA 84:837-841; Kasid *et al.* (1987) Cancer Res. 47:5733-5738; Daly *et al.* (1990) J. Cell Biochem. 43:199-211; Barrett-Lee *et al.* (1990) Br. J Cancer 61:612-617; King *et al.* (1989) J. Steroid Biochem. 34:133-138; Welch *et al.* (1990) Proc. Natl. Acad. Sci. USA 87:7678-7682; Walker *et al.* (1992) Eur. J. Cancer 238:641-644) and induction of TGFß1 by tamoxifen treatment (Butta *et al.* (1992) Cancer Res. 52:4261-4264) has been associated with failure of tamoxifen treatment for breast cancer (Thompson *et al.* (1991) Br. J Cancer 63:609-614). Anti TGFß1 antibodies inhibit the growth of MDA-231 human breast cancer cells in athymic mice (Arteaga *et al.* (1993) J. Clin. Invest. 92:2569-2576), a treatment which is correlated with an increase in spleen natural killer cell activity. CHO cells transfected with latent TGFß1 also showed decreased NK activity and increased tumor growth in nude mice (Wallick *et al.* (1990) J. Exp. Med. 172:1777-1784). Thus, TGFß1 secreted by breast tumors may cause an endocrine immune suppression.

High plasma concentrations of TGFß1 have been shown to indicate poor prognosis for advanced breast cancer patients (Anscher *et al.* (1993) N. Engl. J. Med. 328:1592-1598). Patients with high circulating TGFß before high dose chemotherapy and autologous bone marrow transplantation are at high risk for hepatic veno-occlusive disease (15-50% of all patients with a mortality rate up to 50%) and idiopathic interstitial pneumonitis (40-60% of all patients). The implication of these findings is 1) that elevated plasma levels of TGFß1 can be used to identify at risk patients and 2) that reduction of TGFß1 could decrease the morbidity and mortality of these common treatments for breast cancer patients.

A method for the *in vitro* evolution of nucleic acid molecules with high affinity

binding to target molecules has been developed. This method, Systematic Evolution of Ligands by EXponential enrichment, termed SELEX, is described in United States Patent Application Serial No. 07/536,428, filed June 11, 1990, entitled "Systematic Evolution of Ligands by Exponential Enrichment," now abandoned, United States Patent Application Serial No. 07/714,131, filed June 10, 1991, entitled "Nucleic Acid Ligands," now issued as United States Patent No. 5,475,096, United States Patent Application Serial No. 07/931,473, filed August 17, 1992, entitled "Methods for Identifying Nucleic Acid Ligands," now United States Patent No. 5,270,163 (see also WO91/19813), each of which is herein specifically incorporated by reference. Each of these applications, collectively referred to herein as the SELEX Patent Applications, describe a fundamentally novel method for making a nucleic acid ligand to any desired target molecule.

The SELEX method involves selection from a mixture of candidate oligonucleotides and step-wise iterations of binding, partitioning and amplification, using the same general selection theme, to achieve virtually any desired criterion of binding affinity and selectivity. Starting from a mixture of nucleic acids, preferably comprising a segment of randomized sequence, the SELEX method includes steps of contacting the mixture with the target under conditions favorable for binding, partitioning unbound nucleic acids from those nucleic acids which have bound to target molecules, dissociating the nucleic acid-target complexes, amplifying the nucleic acids dissociated from the nucleic acid-target complexes to yield a ligand-enriched mixture of nucleic acids, then reiterating the steps of binding, partitioning, dissociating and amplifying through as many cycles as desired to yield high affinity nucleic acid ligands to the target molecule.

The basic SELEX method may be modified to achieve specific objectives. For example, United States Patent Application Serial No. 07/960,093, filed October 14, 1992, entitled "Method for Selecting Nucleic Acids on the Basis of Structure," now abandoned, describes the use of SELEX in conjunction with gel electrophoresis to select nucleic acid molecules with specific structural characteristics, such as bent DNA. (See United States Patent No. 5,707,796). United States Patent Application Serial No. 08/123,935, filed September 17, 1993, entitled "Photoselection of Nucleic Acid Ligands," now abandoned, describes a SELEX based method for selecting nucleic acid ligands containing photoreactive groups capable of binding and/or photocrosslinking to and/or photoinactivating a target molecule. (See United States Patent No. 5,763,177). United States Patent Application

Serial No. 08/134,028, filed October 7, 1993, entitled "High-Affinity Nucleic Acid Ligands That Discriminate Between Theophylline and Caffeine," abandoned in favor of United States Patent Application Serial No. 08/443,957, now United States Patent No. 5,580,737, describes a method for identifying highly specific nucleic acid ligands able to discriminate between closely related molecules, termed "Counter-SELEX." United States Patent Application Serial No. 08/143,564, filed October 25, 1993, entitled "Systematic Evolution of Ligands by EXponential Enrichment: Solution SELEX," abandoned in favor of United States Patent Application Serial No. 08/461,061, now United States Patent No. 5,567,588) and United States Patent Application Serial No. 08/792,075, filed January 31, 1997, entitled "Flow Cell SELEX," now United States Patent No. 5,861,254, describe SELEX-based methods which achieve highly efficient partitioning between oligonucleotides having high and low affinity for a target molecule. United States Patent Application Serial No. 07/964,624, filed October 21, 1992, entitled "Nucleic Acid Ligands to HIV-RT and HIV-1 Rev," now United States Patent No. 5,496,938, describes methods for obtaining improved Nucleic Acid Ligands after the SELEX process has been performed. United States Patent Application Serial No. 08/400,440, filed March 8, 1995, entitled "Systematic Evolution of Ligands by EXponential Enrichment: Chemi-SELEX," now United States Patent No. 5,705,337, describes methods for covalently linking a ligand to its target.

The SELEX method encompasses the identification of high-affinity nucleic acid ligands containing modified nucleotides conferring improved characteristics on the ligand, such as improved *in vivo* stability or delivery. Examples of such modifications include chemical substitutions at the ribose and/or phosphate and/or base positions. Specific SELEX-identified nucleic acid ligands containing modified nucleotides are described in United States Patent Application Serial No. 08/117,991, filed September 8, 1993, entitled "High Affinity Nucleic Acid Ligands Containing Modified Nucleotides," abandoned in favor of United States Patent Application Serial No. 08/430,709, now United States Patent No. 5,660,985, that describes oligonucleotides containing nucleotide derivatives chemically modified at the 5- and 2'-positions of pyrimidines, as well as specific RNA ligands to thrombin containing 2'-amino modifications. United States Patent Application Serial No. 08/134,028, *supra*, describes highly specific nucleic acid ligands containing one or more nucleotides modified with 2'-amino (2'-NH_2), 2'-fluoro (2'-F), and/or 2'-O-methyl (2'-OMe). United States Patent Application Serial No. 08/264,029, filed June 22, 1994, entitled

"Novel Method of Preparation of Known and Novel 2' Modified Nucleosides by Intramolecular Nucleophilic Displacement," describes oligonucleotides containing various 2'-modified pyrimidines. International Publication No. WO 98/30720, published July 16, 1998, entitled "Bioconjugation of Oligonucleotides," describes a method for identifying bioconjugates to a target comprising nucleic acid ligands derivatized with a molecular entity exclusively at the 5'-position of the nucleic acid ligands.

The SELEX method encompasses combining selected oligonucleotides with other selected oligonucleotides and non-oligonucleotide functional units as described in United States Patent Application Serial No. 08/284,063, filed August 2, 1994, entitled "Systematic Evolution of Ligands by Exponential Enrichment: Chimeric SELEX," now United States Patent No. 5,637,459 and United States Patent Application Serial No. 08/234,997, filed April 28, 1994, entitled "Systematic Evolution of Ligands by Exponential Enrichment: Blended SELEX," now United States Patent No. 5,683,867, respectively. These applications allow the combination of the broad array of shapes and other properties, and the efficient amplification and replication properties of oligonucleotides with the desirable properties of other molecules. The full text of the above described patent applications, including but not limited to, all definitions and descriptions of the SELEX process, are specifically incorporated herein by reference in their entirety.

BRIEF SUMMARY OF THE INVENTION

The present invention includes methods of identifying and producing nucleic acid ligands to transforming growth factor beta (TGFß) and the nucleic acid ligands so identified and produced. For the purpose of this application, TGFß includes human TGFß1, TGFß2, TGFß3 and TGFß's that are substantially homologous thereto. By substantially homologous it is meant a degree of amino acid sequence identity of 70% or more. In particular, RNA sequences are provided that are capable of binding specifically to TGFß1. Specifically included in the invention are the RNA ligand sequences shown in Table 3 (SEQ ID NOS:6-143). Also included in this invention are RNA ligands of TGFß1 that inhibit the function of TGFß1.

Further included in this invention is a method of identifying nucleic acid ligands and nucleic acid ligand sequences to TGFß comprising the steps of (a) preparing a candidate mixture of nucleic acids, (b) contacting the candidate mixture of nucleic acids with TGFß,

(c) partitioning between members of said candidate mixture on the basis of affinity to TGFß, and (d) amplifying the selected molecules to yield a mixture of nucleic acids enriched for nucleic acid sequences with a relatively higher affinity for binding to TGFß.

More specifically, the present invention includes the RNA ligands to TGFß identified according to the above-described method, including those ligands shown in Table 3 (SEQ ID NOS:6-143). Also included are nucleic acid ligands to TGFß that are substantially homologous to any of the given ligands and that have substantially the same ability to bind TGFß and inhibit the function of TGFß. Further included in this invention are nucleic acid ligands to TGFß that have substantially the same structural form as the ligands presented herein and that have substantially the same ability to bind TGFß and inhibit the function of TGFß.

The present invention also includes other modified nucleotide sequences based on the nucleic acid ligands identified herein and mixtures of the same.

BRIEF DESCRIPTION OF THE FIGURES

Figures 1A and 1B show the binding curves of rounds 0 (○), 14 (▲), 15 (■) and 16 (●) of the 40N pool (Fig. 1A) and rounds 0 (○), 14 (■), 15 (▲) and 17 (●) of the 30N pool (Fig. 1B) presented as %RNA bound vs. concentration of TGFß1.

Figure 2 shows the affinity sensorgram of random RNA (◊), ligand 40-03 (○), ligand 40-60 (▲) and polyclonal anti TGFß1 antibody (●) performed on TGFß1, expressed as response units vs. time.

Figures 3A-3C show sensorgrams obtained in a binding specificity analysis of TGFß1 performed on random RNA (Fig. 3A), ligand 40-03 (Fig. 3B) and ligand 40-60 (Fig. 3C) with various concentrations of TGFß1, expressed as response units vs. time. Figures 3D-3F show sensorgrams obtained in a binding specificity analysis of TGFß2 performed on random RNA (Fig. 3D), ligand 40-03 (Fig. 3E) and ligand 40-60 (Fig. 3F) with various concentrations of TGFß2, expressed as response units vs. time.

Figures 4A and 4B illustrate the results of the TGFß1 bioasay on mink lung epithelial cells (MLEC). Figures 4A and 4B show the inhibitory activity of rounds 11 (■) and 14 (●) of the 40N pool (Fig. 4A) and rounds 11 (■) and 14 (●) of the 30N pool (Fig. 4B) compared to random RNA (▲). The results are expressed as ^{3}H-thymidine incorporation as

net % of control vs. concentration of TGFß1, where control is the amount of ^{3}H-thymidine incorporation in the absence of TGFß1 and RNA minus the amount of incorporation in the presence of TGFß1 alone.

Figures 5A-5D illustrate the results of the TGFß1 bioassay on mink lung epithelial cells (MLEC). Figure 5A is a TGFß1 titration curve presented as ^{3}H-thymidine incorporation as a per cent of control vs. concentration of TGFß1. Figures 5B-5D illustrate the bioactivities of round 16 of the 40N pool (Fig. 5B, (●)), ligand 40-03 (Fig. 5C, (●)) and ligand 40-60 (Fig. 5D, (●)) as compared to the bioactivities of a polyclonal anti-TGFß1 antibody (○) and random RNA (■), presented as ^{3}H-thymidine incorporation as a per cent of control vs. concentration of TGFß1.

Figure 6 shows the bioactivities of random RNA (■), ligand 40-60 (▲), ligand 40-03 (●), a monoclonal antibody specific for TGFß2 and TGFß3 (○) and a pan-specific antibody specific for TGFß1, TGFß2 and TGFß3 (Δ), presented as ^{3}H-thymidine incorporation as a per cent of control vs. concentration of TGFß1.

Figure 7 is a proposed folding of the class 1 bioactive ligands. S1, S2 and S3 designate stem 1, stem 2 and stem 3 of the proposed structure.

DETAILED DESCRIPTION OF THE INVENTION

This application describes high-affinity nucleic acid ligands to TGFß identified through the method known as SELEX. SELEX is described in United States Patent Application Serial No. 07/536,428, entitled "Systematic Evolution of Ligands by EXponential Enrichment," now abandoned, United States Patent Application Serial No. 07/714,131, filed June 10, 1991, entitled "Nucleic Acid Ligands," now United States Patent No. 5,475,096, United States Patent Application Serial No. 07/931,473, filed August 17, 1992, entitled "Methods for Identifying Nucleic Acid Ligands," now United States Patent No. 5,270,163, (see also WO91/19813). These applications, each specifically incorporated herein by reference, are collectively called the SELEX Patent Applications. Certain terms used to described the invention herein are defined as follows.

"Nucleic Acid Ligand" as used herein is a non-naturally occurring nucleic acid having a desirable action on a target. A desirable action includes, but is not limited to, binding of the target, catalytically changing the target, reacting with the target in a way which modifies/alters the target or the functional activity of the target, covalently attaching

to the target as in a suicide inhibitor, and facilitating the reaction between the target and another molecule. In the preferred embodiment, the desirable action is specific binding to a target molecule, such target molecule being a three dimensional chemical structure other than a polynucleotide that binds to the nucleic acid ligand through a mechanism which predominantly depends on Watson/Crick base pairing or triple helix binding, wherein the nucleic acid ligand is not a nucleic acid having the known physiological function of being bound by the target molecule. Nucleic acid ligands include nucleic acids that are identified from a candidate mixture of nucleic acids, said nucleic acid ligand being a ligand of a given target by the method comprising: a) contacting the candidate mixture with the target, wherein nucleic acids having an increased affinity to the target relative to the candidate mixture may be partitioned from the remainder of the candidate mixture; b) partitioning the increased affinity nucleic acids from the remainder of the candidate mixture; and c) amplifying the increased affinity nucleic acids to yield a ligand-enriched mixture of nucleic acids.

"Candidate Mixture" is a mixture of nucleic acids of differing sequence from which to select a desired ligand. The source of a candidate mixture can be from naturally-occurring nucleic acids or fragments thereof, chemically synthesized nucleic acids, enzymatically synthesized nucleic acids or nucleic acids made by a combination of the foregoing techniques. In a preferred embodiment, each nucleic acid has fixed sequences surrounding a randomized region to facilitate the amplification process.

"Nucleic Acid" means either DNA, RNA, single-stranded or double-stranded and any chemical modifications thereof. Modifications include, but are not limited to, those which provide other chemical groups that incorporate additional charge, polarizability, hydrogen bonding, electrostatic interaction, and fluxionality to the nucleic acid ligand bases or to the nucleic acid ligand as a whole. Such modifications include, but are not limited to, 2'-position sugar modifications, 5-position pyrimidine modifications, 8-position purine modifications, modifications at exocyclic amines, substitution of 4-thiouridine, substitution of 5-bromo or 5-iodo-uracil, backbone modifications, methylations, unusual base-pairing combinations such as the isobases isocytidine and isoguanidine and the like. Modifications can also include 3' and 5' modifications such as capping.

"SELEX" methodology involves the combination of selection of nucleic acid ligands which interact with a target in a desirable manner, for example binding to a protein, with

amplification of those selected nucleic acids. Iterative cycling of the selection/amplification steps allows selection of one or a small number of nucleic acids which interact most strongly with the target from a pool which contains a very large number of nucleic acids. Cycling of the selection/amplification procedure is continued until a selected goal is achieved. In the present invention, the SELEX methodology is employed to obtain nucleic acid ligands to TGFß. The SELEX methodology is described in the SELEX Patent Applications.

"Target" means any compound or molecule of interest for which a ligand is desired. A target can be a protein, peptide, carbohydrate, polysaccharide, glycoprotein, hormone, receptor, antigen, antibody, virus, substrate, metabolite, transition state analog, cofactor, inhibitor, drug, dye, nutrient, growth factor, etc. without limitation. In this application, the target is a TGFß, preferably TGFß1.

In its most basic form, the SELEX process may be defined by the following series of steps.

1) A candidate mixture of nucleic acids of differing sequence is prepared. The candidate mixture generally includes regions of fixed sequences (i.e., each of the members of the candidate mixture contains the same sequences in the same location) and regions of randomized sequences. The fixed sequence regions are selected either: (a) to assist in the amplification steps described below, (b) to mimic a sequence known to bind to the target, or (c) to enhance the concentration of a given structural arrangement of the nucleic acids in the candidate mixture. The randomized sequences can be totally randomized (i.e., the probability of finding a base at any position being one in four) or only partially randomized (e.g., the probability of finding a base at any location can be selected at any level between 0 and 100 percent).

2) The candidate mixture is contacted with the selected target under conditions favorable for binding between the target and members of the candidate mixture. Under these circumstances, the interaction between the target and the nucleic acids of the candidate mixture can be considered as forming nucleic acid-target pairs between the target and those nucleic acids having the strongest affinity for the target.

3) The nucleic acids with the highest affinity for the target are partitioned from those nucleic acids with lesser affinity to the target. Because only an extremely small number of sequences (and possibly only one molecule of nucleic acid) corresponding to the highest affinity nucleic acids exist in the candidate mixture, it is generally desirable to set the

partitioning criteria so that a significant amount of the nucleic acids in the candidate mixture (approximately 5-50%) are retained during partitioning.

4) Those nucleic acids selected during partitioning as having the relatively higher affinity to the target are then amplified to create a new candidate mixture that is enriched in nucleic acids having a relatively higher affinity for the target.

5) By repeating the partitioning and amplifying steps above, the newly formed candidate mixture contains fewer and fewer weakly binding sequences, and the average degree of affinity of the nucleic acids to the target will generally increase. Taken to its extreme, the SELEX process will yield a candidate mixture containing one or a small number of unique nucleic acids representing those nucleic acids from the original candidate mixture having the highest affinity to the target molecule.

The SELEX Patent Applications describe and elaborate on this process in great detail. Included are targets that can be used in the process; methods for partitioning nucleic acids within a candidate mixture; and methods for amplifying partitioned nucleic acids to generate enriched candidate mixture. The SELEX Patent Applications also describe ligands obtained to a number of target species, including both protein targets where the protein is and is not a nucleic acid binding protein.

The SELEX method further encompasses combining selected nucleic acid ligands with lipophilic or non-immunogenic, high molecular weight compounds in a diagnostic or therapeutic complex as described in United States Patent Application No. 08/434,465, filed May 4, 1995, entitled "Nucleic Acid Ligand Complexes." VEGF nucleic acid ligands that are associated with a lipophilic compound, such as diacyl glycerol or dialkyl glycerol, in a diagnostic or therapeutic complex are described in United States Patent Application Serial No. 08/739,109, filed October 25, 1996, entitled "Vascular Endothelial Growth Factor (VEGF) Nucleic Acid Ligand Complexes," now United States Patent No. 5,859,228. VEGF nucleic acid ligands that are associated with a lipophilic compound, such as a glycerol lipid, or a non-immunogenic, high molecular weight compound, such as polyalkylene glycol, are further described in United States Patent Application Serial No. 08/897,351, filed July 21, 1997, entitled "Vascular Endothelial Growth Factor (VEGF) Nucleic Acid Ligand Complexes." VEGF nucleic acid ligands that are associated with a non-immunogenic, high molecular weight compound or lipophilic compound are also further described in WO 98/18480, published May 7, 1998, entitled "Vascular Endothelial Growth Factor (VEGF) Nucleic Acid

Ligand Complexes." Each of the above described patent applications which describe modifications of the basic SELEX procedure are specifically incorporated by reference herein in their entirety.

Certain embodiments of the present invention provide a complex comprising one or more nucleic acid ligands to TGFß covalently linked with a non-immunogenic, high molecular weight compound or lipophilic compound. A complex as used herein describes the molecular entity formed by the covalent linking of the nucleic acid ligand of TGFß to a non-immunogenic, high molecular weight compound. A non-immunogenic, high molecular weight compound is a compound between approximately 100 Da to 1,000,000 Da, more preferably approximately 1000 Da to 500,000 Da, and most preferably approximately 1000 Da to 200,000 Da, that typically does not generate an immunogenic response. For the purposes of this invention, an immunogenic response is one that causes the organism to make antibody proteins. In a preferred embodiment of the invention, the non-immunogenic, high molecular weight compound is a polyalkylene glycol. In the most preferred embodiment, the polyalkylene glycol is polyethylene glycol (PEG). More preferably, the PEG has a molecular weight of about 10-80K. Most preferably, the PEG has a molecular weight of about 20-45K. In certain embodiments of the invention, the non-immunogenic, high molecular weight compound can also be a nucleic acid ligand.

Another embodiment of the invention is directed to complexes comprised of a nucleic acid ligand to TGFß and a lipophilic compound. Lipophilic compounds are compounds that have the propensity to associate with or partition into lipids and/or other materials or phases with low dielectric constants, including structures that are comprised substantially of lipophilic components. Lipophilic compounds include lipids as well as non-lipid containing compounds that have the propensity to associate with lipids (and/or other materials or phases with low dielectric constants). Cholesterol, phospholipid and glycerol lipids, such as dialkylglycerol, diacylglycerol, and glycerol amide lipids are further examples of lipophilic compounds. In a preferred embodiment, the lipophilic compound is a glycerol lipid.

The non-immunogenic, high molecular weight compound or lipophilic compound may be covalently bound to a variety of positions on the nucleic acid ligand to TGFß, such as to an exocyclic amino group on the base, the 5-position of a pyrimidine nucleotide, the 8-position of a purine nucleotide, the hydroxyl group of the phosphate, or a hydroxyl group or other group at the 5' or 3' terminus of the nucleic acid ligand to TGFß. In embodiments where the lipophilic

compound is a glycerol lipid, or the non-immunogenic, high molecular weight compound is polyalkylene glycol or polyethylene glycol, preferably the non-immunogenic, high molecular weight compound is bonded to the 5' or 3' hydroxyl of the phosphate group thereof. In the most preferred embodiment, the lipophilic compound or non-immunogenic, high molecular weight compound is bonded to the 5' hydroxyl of the phosphate group of the nucleic acid ligand. Attachment of the non-immunogenic, high molecular weight compound or lipophilic compound to the nucleic acid ligand of TGFß can be done directly or with the utilization of linkers or spacers.

A linker is a molecular entity that connects two or more molecular entities through covalent bonds or non-covalent interactions, and can allow spatial separation of the molecular entities in a manner that preserves the functional properties of one or more of the molecular entities. A linker can also be referred to as a spacer.

The complex comprising a nucleic acid ligand to TGFß and a non-immunogenic, high molecular weight compound or lipophilic compound can be further associated with a lipid construct. Lipid constructs are structures containing lipids, phospholipids, or derivatives thereof comprising a variety of different structural arrangements which lipids are known to adopt in aqueous suspension. These structures include, but are not limited to, lipid bilayer vesicles, micelles, liposomes, emulsions, lipid ribbons or sheets, and may be complexed with a variety of drugs and components which are known to be pharmaceutically acceptable. In the preferred embodiment, the lipid construct is a liposome. The preferred liposome is unilamellar and has a relative size less than 200 nm. Common additional components in lipid constructs include cholesterol and alpha-tocopherol, among others. The lipid constructs may be used alone or in any combination which one skilled in the art would appreciate to provide the characteristics desired for a particular application. In addition, the technical aspects of lipid constructs and liposome formation are well known in the art and any of the methods commonly practiced in the field may be used for the present invention.

The SELEX method further comprises identifying bioconjugates to a target. Copending International Publication No. WO 98/30720, published July 6, 1998, entitled "Bioconjugation of Oligonucleotides," describes a method for enzymatically synthesizing bioconjugates comprising RNA derivatized exclusively at the 5'-position with a molecular entity, and a method for identifying bioconjugates to a target comprising nucleic acid ligands derivatized with a molecular entity exclusively at the 5'-position of the nucleic acid

ligands. A bioconjugate as used herein refers to any oligonucleotide which has been derivatized with another molecular entity. In the preferred embodiment, the molecular entity is a macromolecule. As used herein, a macromolecule refers to a large organic molecule. Examples of macromolecules include, but are not limited to nucleic acids, oligonucleotides, proteins, peptides, carbohydrates, polysaccharides, glycoproteins, lipophilic compounds, such as cholesterol, phospholipids, diacyl glycerols and dialkyl glycerols, hormones, drugs, non-immunogenic high molecular weight compounds, fluorescent, chemiluminescent and bioluminescent marker compounds, antibodies and biotin, etc. without limitation. In certain embodiments, the molecular entity may provide certain desirable characteristics to the nucleic acid ligand, such as increasing RNA hydrophobicity and enhancing binding, membrane partitioning and/or permeability. Additionally, reporter molecules, such as biotin, fluorescein or peptidyl metal chelates for incorporation of diagnostic radionuclides may be added, thus providing a bioconjugate which may be used as a diagnostic agent.

Certain embodiments of the present invention provide bioconjugates to TGFß comprising RNA derivatized exclusively at the 5'-position with a molecular entity obtained by the enzymatic method described in WO 98/30720. Other embodiments of the present invention provide bioconjugates to TGFß comprising a nucleic acid ligand covalently bonded to a macromolecule, obtained from a candidate mixture of bioconjugates, obtained by the method described in WO 98/30720.

The methods described herein and the nucleic acid ligands identified by such methods are useful for both therapeutic and diagnostic purposes. Therapeutic uses include the treatment or prevention of diseases or medical conditions in human patients. Therapeutic uses may also include veterinary applications.

Diagnostic utilization may include both *in vivo* or *in vitro* diagnostic applications. The SELEX method generally, and the specific adaptations of the SELEX method taught and claimed herein specifically, are particularly suited for diagnostic applications. SELEX identifies nucleic acid ligands that are able to bind targets with high affinity and with surprising specificity. These characteristics are, of course, the desired properties one skilled in the art would seek in a diagnostic ligand.

The nucleic acid ligands of the present invention may be routinely adapted for diagnostic purposes according to any number of techniques employed by those skilled in the

art or by the methods described in WO 98/30720, *supra*. Diagnostic agents need only be able to allow the user to identify the presence of a given target at a particular locale or concentration. Simply the ability to form binding pairs with the target may be sufficient to trigger a positive signal for diagnostic purposes. Those skilled in the art would also be able to adapt any nucleic acid ligand by procedures known in the art to incorporate a labeling tag in order to track the presence of such ligand. Such a tag could be used in a number of diagnostic procedures. The nucleic acid ligands to TGFß described herein may specifically be used for identification of the TGFß protein.

SELEX provides high affinity ligands of a target molecule. This represents a singular achievement that is unprecedented in the field of nucleic acids research. The present invention applies the SELEX procedure to the specific target of TGFß1. In the Example section below, the experimental parameters used to isolate and identify the nucleic acid ligands to TGFß1 are described.

In order to produce nucleic acids desirable for use as a pharmaceutical, it is preferred that the nucleic acid ligand (1) binds to the target in a manner capable of achieving the desired effect on the target; (2) be as small as possible to obtain the desired effect; (3) be as stable as possible; and (4) be a specific ligand to the chosen target. In most situations, it is preferred that the nucleic acid ligand have the highest possible affinity to the target.

In the present invention, SELEX experiments were performed in order to identify RNA ligands with specific high affinity for TGFß1 from degenerate libraries containing 20, 30 or 40 random positions (20N7 (SEQ ID NO:1), 30N7 (SEQ ID NO:2) or 40N7 (SEQ ID NO:3)) (Table 1). This invention includes the specific RNA ligands to TGFß1 shown in Table 3 (SEQ ID NOS:6-143), identified by the methods described in Examples 1 and 2. This invention further includes RNA ligands to TGFß1 which inhibit TGFß1 function, presumably by inhibiting the interaction of TGFß1 with its receptor. The scope of the ligands covered by this invention extends to all nucleic acid ligands of TGFß, modified and unmodified, identified according to the SELEX procedure. More specifically, this invention includes nucleic acid sequences that are substantially homologous to the ligands shown in Table 3 (SEQ ID NOS:6-143). By substantially homologous it is meant a degree of primary sequence homology in excess of 70%, most preferably in excess of 80%, and even more preferably in excess of 90%, 95% or 99%. The percentage of homology as described herein is calculated as the percentage of nucleotides found in the smaller of the two sequences

which align with identical nucleotide residues in the sequence being compared when 1 gap in a length of 10 nucleotides may be introduced to assist in that alignment. A review of the sequence homologies of the ligands of TGFß shown in Tables 3 (SEQ ID NOS:6-143) shows that some sequences with little or no primary homology may have substantially the same ability to bind TGFß. For this reason, this invention also includes nucleic acid ligands that have substantially the same structure and ability to bind TGFß as the nucleic acid ligands shown in Table 3 (SEQ ID NOS:6-143). Substantially the same ability to bind TGFß means that the affinity is within one or two orders of magnitude of the affinity of the ligands described herein. It is well within the skill of those of ordinary skill in the art to determine whether a given sequence -- substantially homologous to those specifically described herein -- has substantially the same ability to bind TGFß.

This invention also includes nucleic acid ligands that have substantially the same postulated structure or structural motifs. Substantially the same structure or structural motifs can be postulated by sequence alignment using the Zukerfold program (see Zuker (1989) Science 244:48-52) as would be known in the art, other computer programs can be used for predicting secondary structure and structural motifs. Substantially the same structure or structural motif of nucleic acid ligands in solution or as a bound structure can also be postulated using NMR or other techniques as would be known in the art.

One potential problem encountered in the therapeutic, prophylactic, and *in vivo* diagnostic use of nucleic acids is that oligonucleotides in their phosphodiester form may be quickly degraded in body fluids by intracellular and extracellular enzymes such as endonucleases and exonucleases before the desired effect is manifest. Certain chemical modifications of the nucleic acid ligand can be made to increase the *in vivo* stability of the nucleic acid ligand or to enhance or to mediate the delivery of the nucleic acid ligand. See, e.g., United States Patent Application Serial No. 08/117,991, filed September 8, 1993, entitled "High Affinity Nucleic Acid Ligands Containing Modified Nucleotides," abandoned in favor of United States Patent Application Serial No. 08/430,709, now issued as United States Patent No. 5,660,985 and United States Patent Application Serial No. 08/434,465, filed May 4, 1995, entitled "Nucleic Acid Ligand Complexes," which are specifically incorporated herein by reference. Modifications of the nucleic acid ligands contemplated in this invention include, but are not limited to, those which provide other chemical groups that incorporate additional charge, polarizability, hydrophobicity, hydrogen bonding,

electrostatic interaction, and fluxionality to the nucleic acid ligand bases or to the nucleic acid ligand as a whole. Such modifications include, but are not limited to, 2'-position sugar modifications, 5-position pyrimidine modifications, 8-position purine modifications, modifications at exocyclic amines, substitution of 4-thiouridine, substitution of 5-bromo or 5-iodo-uracil, backbone modifications, phosphorothioate or alkyl phosphate modifications, methylations, unusual base-pairing combinations such as the isobases isocytidine and isoguanidine and the like. Modifications can also include 3' and 5' modifications such as capping.

Where the nucleic acid ligands are derived by the SELEX method, the modifications can be pre- or post- SELEX modifications. Pre-SELEX modifications yield nucleic acid ligands with both specificity for their SELEX target and improved *in vivo* stability. Post-SELEX modifications made to 2'-OH nucleic acid ligands can result in improved *in vivo* stability without adversely affecting the binding capacity of the nucleic acid ligand. The preferred modifications of the nucleic acid ligands of the subject invention are 5' and 3' phosphorothioate capping and/or 3'-3' inverted phosphodiester linkage at the 3' end. In one preferred embodiment, the preferred modification of the nucleic acid ligand is a 3'-3' inverted phosphodiester linkage at the 3' end. Additional 2'-fluoro (2'-F) and/or 2'-amino (2'-NH_2) and/or 2'-O methyl (2'-OMe) modification of some or all of the nucleotides is preferred. Described herein are nucleic acid ligands that were 2'-F modified and incorporated into the SELEX process. Other modifications are known to one of ordinary skill in the art. Such modifications may be made post-SELEX (modification of previously identified unmodified ligands) or by incorporation into the SELEX process.

As described above, because of their ability to selectively bind TGFß, the nucleic acid ligands to TGFß described herein are useful as pharmaceuticals. This invention, therefore, also includes a method for treating TGFß-mediated pathological conditions by administration of a nucleic acid ligand capable of binding to TGFß.

Therapeutic compositions of the nucleic acid ligands may be administered parenterally by injection, although other effective administration forms, such as intraarticular injection, inhalant mists, orally active formulations, transdermal iontophoresis or suppositories, are also envisioned. One preferred carrier is physiological saline solution, but it is contemplated that other pharmaceutically acceptable carriers may also be used. In one preferred embodiment, it is envisioned that the carrier and the ligand constitute a

physiologically-compatible, slow release formulation. The primary solvent in such a carrier may be either aqueous or non-aqueous in nature. In addition, the carrier may contain other pharmacologically-acceptable excipients for modifying or maintaining the pH, osmolarity, viscosity, clarity, color, sterility, stability, rate of dissolution, or odor of the formulation. Similarly, the carrier may contain still other pharmacologically-acceptable excipients for modifying or maintaining the stability, rate of dissolution, release, or absorption of the ligand. Such excipients are those substances usually and customarily employed to formulate dosages for parental administration in either unit dose or multi-dose form.

Once the therapeutic composition has been formulated, it may be stored in sterile vials as a solution, suspension, gel, emulsion, solid, or dehydrated or lyophilized powder. Such formulations may be stored either in a ready to use form or requiring reconstitution immediately prior to administration. The manner of administering formulations containing nucleic acid ligands for systemic delivery may be via subcutaneous, intramuscular, intravenous, intranasal or vaginal or rectal suppository.

The following Examples are provided to explain and illustrate the present invention and are not intended to be limiting of the invention. Example 1 describes the various materials and experimental procedures used in Example 2. Example 2 describes a representative method for identifying RNA ligands by the SELEX method which bind TGFß1. Example 3 describes the affinities the ligands have for TGFß1. Example 4 describes the specificity of ligands to hTGFß1. Example 5 describes the inhibition of TGFß1 bioactivity with several ligands. Example 6 summarizes the results of the data from Examples 2-5. Example 7 describes the proposed secondary structure of bioactive TGFß1 ligands.

EXAMPLES

Example 1. Experimental Procedures

a) Materials

Recombinant human Transforming Growth Factor Beta 1 (hTGFß1) was purchased from R&D Systems (Minneapolis, MN). Mink Lung Epithelial Cells (MLEC) were obtained from American Type Culture Collection (MV 1 Lu ATCC No. CCL 64). T7 RNA polymerase, 2'-F-modified CTP and UTP were prepared in house. DNA oligonucleotides

were obtained from Operon Technologies, Inc. (Alameda, CA). All other reagents and chemicals were from commercial sources.

b) SELEX

The SELEX process has been described in detail in United States Patent No. 5,270,163 (see also Tuerk and Gold (1990) Science 249:505-510). The DNA templates contained either 40 (SEQ ID NO:1), 30 (SEQ ID NO:2) or 20 (SEQ ID NO:3) random nucleotides, flanked by 5' and 3' constant regions for primer annealing sites for PCR and cDNA synthesis (Table 1). The starting pool of single stranded DNA molecules were converted to double stranded DNA by primer extension reactions with the klenow fragment of DNA polymerase. RNA pools were prepared by transcription and were gel purified before use. Transcription reactions were done with about 5 μM DNA template, 5 units/μL T7 RNA polymerase, 40 mM Tris-HCl (pH 8), 12 mM $MgCl_2$, 5 mM DTT, 1 mM spermidine, 0.002% Triton X-100, 4% PEG 8000, 2-4 mM each 2'-OH ATP, 2'-OH GTP, 2'-F CTP, 2'-F UTP, and 0.25 μM α-^{32}P-2'-OH ATP (800 Ci/mmole). At later rounds, RNA pools were prefiltered and/or preadsorbed with multiple layers of the same nitrocellulose filter type used in the SELEX process in order to reduce the frequency of molecules selected for nitrocellulose binding. To prepare binding reactions, the RNA molecules were incubated with recombinant hTGFß1 in Dulbecco's Phosphate-Buffered Saline (DPBS) (Life Technologies, Gaithersburg, MD, Cat. No 21600-010) containing 0.01% human serum albumin and 1.0 mM $MgCl_2$. Following incubation at 37°C (10 minutes to 10 hours) the protein-RNA complexes were partitioned from unbound RNA by capture on nitrocellulose. Nitrocellulose filter bound RNA was recovered by phenol/urea extraction. The partitioned RNA was reverse transcribed into cDNA by AMV reverse transcriptase at 48°C for 60 minutes in 50 mM Tris-HCl pH 8.3, 60 mM NaCl, 6 mM $Mg(OAc)_2$, 10 mM DTT, 50 pmol DNA 3' primer 3G7 (SEQ ID NO:5; Table 1), 0.4 mM each of dATP, dCTP, dGTP, and dTTP, and 1 unit/μL AMV RT. The cDNA was PCR amplified and used to initiate the next SELEX cycle. PCR conditions were 2 μM each 3G7 (SEQ ID NO:5) and 5G7 (SEQ ID NO:4) primers (Table 1), 50 mM KCl, 10 mM Tris-HCl, pH 9, 0.1% Triton X-100, 3 mM $MgCl_2$, 0.5 mM of each dATP, dCTP, dGTP, and dTTP, and 0.1 units/μL Taq DNA polymerase.

c) Nitrocellulose Filter Partitioning

To partition the protein-RNA complexes away from uncomplexed RNA, the binding reactions were filtered through nitrocellulose/cellulose acetate mixed matrix, 0.45 μm pore size filter disks, type HA, (Millipore, Co., Bedford, MA). For filtration, the filters were placed onto a vacuum manifold and wetted by aspirating with 5 mL of DPBS. The binding reactions were aspirated through the filters, washed with 5 mL of DPBS + $MgCl_2$ and counted in a scintillation counter (Beckmann). At later rounds, nitrocellulose filters were preblocked with 2 mL of DPBS + 1 mM $MgCl_2$ + 0.01% BSA, and wash volumes were increased to 25 mL in order to reduce background binding to nitrocellulose. At later rounds in the SELEX process, 10 mL washes with 0.5 M urea were introduced to remove RNA that binds to nitrocellulose.

Nitrocellulose partitioning was also used for determining the equilibrium dissociation constants of RNA ligands to hTGFß1. Binding curves obtained by nitrocellulose filtration indicated that RNA pools and some RNA ligands bind monophasically while others bind biphasically. Biphasic binding can be described as the binding of two affinity species derived from the same ligand sequence that can fold into alternate structures which are kinetically trapped and are not in equilibrium.

To obtain the equilibrium dissociation constants of RNA ligands to TGFß1, the binding reaction:

$$R:P \underset{}{\overset{K_D}{\rightleftharpoons}} R+P$$

where R=RNA, P=Protein and K_D=dissociation constant is converted into an equation for the fraction of RNA bound at equilibrium:

$$q=(f/2R_T)(P_T+R_T+K_D-((P_T+R_T+K_D)^2-4P_TR_T)^{1/2})$$

where q=fraction of RNA bound, P_T=total protein concentration, R_T=total RNA concentration and f=retention efficiency of RNA-protein complexes. The average retention efficiency for RNA- hTGFß1 complexes on nitrocellulose filters is 0.4-0.8.

Biphasic binding data were evaluated using the equation:

$$q = 2P_T + R_T + K_{D1} + K_{D2} - [(P_T + X_1R_1 + K_{D1})^2 - 4P_TX_1R_T]^{1/2} - [(P_T + X_2R_T + K_{D2})^2 - 4P_TX_2R_T]^{1/2}$$

where X_1 and X_2 are the mole fractions of the affinity species R_1 and R_2 and K_{D1} and K_{D2} are the corresponding dissociation constants.

The K_D's were determined by least square fitting of the data points using the software Kaleidagraph (Synergy Software, Reading, PA).

d) Cloning and Sequencing

RNA recovered from the filters of the final round of the SELEX process was reverse transcribed and PCR amplified as in previous rounds. The PCR products were purified by PAG electrophoresis and cloned into the SrfI restriction site of PCR-Script Direct SK(+) plasmid using the pCR-Script Amp SK(+) cloning kit (STRATAGENE CLONING SYSTEMS, La Jolla, CA). About 180 clones were sequenced with ABI Prism sequencing kit (Applied Biosystems, Perkin-Elmer, CT).

e) Analysis of nucleic acid ligand binding by BIAcore

Biotinylated TGFß1 (catalog No. NFTG0, R&D Systems, Minneapolis, MN) was coupled onto an SA5 streptavidin BIAcore chip (BIAcore, Inc., Piscataway, NJ) by injecting biotinylated TGFß1 solution as prepared per manufacturers instructions at 5 µL/min to achieve loadings of 436, 133 and 57 response units (RU) in flow cells 1, 2 and 3, respectively. Flow cell 4 was kept blank for control and background subtractions. To measure binding activities, RNA ligands and antiserum (pan-specific anti-TGFß1 total rabbit IgG, catalog No. AB-100-NA, R&D Systems, Minneapolis, MN) were injected at various concentrations in HBSMC-HSA (Hepes buffered saline pH 7.5, 1 mM $MgCl_2$, 1 mM $CaCl_2$, 0.01% human serum albumin) at 20 µL/min. Injections allowed about 3 minute association and 3 minute dissociation cycles. Data were plotted and analyzed by Bianalysis software (BIAcore, Inc., Piscataway, NJ).

f) Analysis of nucleic acid ligand specificity by BIAcore

Biotinylated 2'-fluoro-pyrimidine RNA nucleic acid ligands were transcribed in the presence of 5'- biotin-modified guanosine monophosphate (5'-biotin-GAP) as described in copending International Publication No. WO 98/30720, published July 6, 1998, the contents of which are incorporated herein by reference. Typical reactions were 1 mL in volume containing standard T7 RNA polymerase, 40 mM Tris-HCl (pH 8), 12 mM $MgCl_2$, 5mM DTT, 1 mM spermidine, 0.002% Triton X-100, 4% PEG 8000, with 3 mM each 2'-F-CTP and 2'-F-UTP, and 1 mM each ATP and GTP and 5 mM 5'-biotin GAP. Following overnight incubation at 37°C, transcripts were purified by gel electrophoresis and ethanol precipitation.

To prepare an analysis chip, three RNA species were used and were injected in HBSMC-HSA at 5 μL/min. Flow-cells 1, 2 and 3 were loaded with 535, 536 and 563 RU of random 40N7 library, TGFß1 ligand 40-03 (SEQ ID NO:84), and TGFß1 ligand 40-60 (SEQ ID NO:128), respectively. Thus, for stoichiometric binding of RNA to TGFß1 or TGFß2, one would expect a maximum of approximately 500 RU's, since TGFß1 and TGFß2 have the same mass as the RNA. Flow cell 4 was kept blank for control and background subtractions. The analysis chip was exposed to various concentrations of TGFß1 and TGFß2 at 20 μL/min. in HBSMC-HSA. Data were plotted and analyzed by Bianalysis software (BIAcore, Inc., Piscataway, NJ).

g) Inhibition of TGFß1 mediated growth suppression of mink lung epithelial cells (MLEC)

To determine the bioactivity of RNA pools and individual ligands, a growth assay was used in which TGFß1 antagonists cause reversal of TGFß1 growth suppression of mink lung epithelial cells. In this assay, MLEC were treated with various concentrations of random RNA, individual ligands, antibodies such as polyclonal anti-TGFß1 antibody (pan-specific anti-TGFß1 total rabbint IgG, catalog No. AB-100-NA, R&D Systems, Minneapolis, MN), monoclonal mouse anti-TGFß2/TGFß3 antibody (Genzyme Corp., Cambridge MA, catalog No. 1836-01) and monoclonal mouse anti-TGFß1/ TGFß2/ hTGFß3 antibody (Genzyme Corp., Cambridge, MA, catalog No. 1835-01) in serum-free 48 hr-3T3-conditioned medium (CM).

Cells were plated at 10^5/mL in 96-well plates in MEM, 10 mM HEPES and 0.2% FBS. Following 4 hours of incubation at 37°C, when cells appeared to attach to the well surface, TGFß1 was added at 2 pM with or without TGFß1 ligands that ranged from 0.1 nM to 1 μM. In a second assay performed in order to determine cross-species reactivity, rather than using hTGFß, a conditioned serum-free medium (CM) was used. CM was conditioned by culturing it in murine 3T3 fibroblast for 48 hours. Before use, this conditioned medium was heat treated at 80°C for 10 minutes to activate the 3T3 cell derived TGFß and then it was diluted to 50% and supplemented with 0.2% murine serum. In each assay, hTGFß1 (or CM) was diluted appropriately in MEM and FBS (0.2% or murine serum) and the ligands were diluted in MEM. TGFß1 (or CM) and ligand dilutions at 10X the final concentration were premixed at equal volumes and then were added to the cells. Following addition of the TGFß1 (or CM) -ligand mixture, the cells were incubated for 16-18 hours prior to addition

of ^{3}H-thymidine at 0.25 μCi per well and continued incubation for 7-8 additional hours. After incubation, the cells were washed and harvested with SKATRON filtering units and ^{3}H-thymidine incorporation in cellular DNA was quantitated by scintillation counting in Ecoscint. Data were plotted and analyzed as described in Park *et al.* (1990) J. Exp. Med. 171:1073) and Dower *et al.* (1984) J. Immunol. 132:751). K_i values were determined from inhibition IC_{50} values according to the equation $K_i=IC_{50}/(1+([T]/K_{dT})$, where [T] is the concentration in molar of TGFß1 present in the assay and K_{dT} is the concentration of TGFß1 causing 50% inhibition of MLEC proliferation as determined by TGFß1 titration experiments.

Example 2. RNA ligands to hTGFß1

a) TGFß1 SELEX

Three parallel SELEX processes were performed with 2'-F pyrimidine modified RNA randomized at 40, 30 and 20 contiguous positions. The conditions for the SELEX process and results for each round are summarized in Table 2. The first round was done under two different conditions where RNA to protein ratios were 10:1 and 50:1. Each condition included a pool of $1.2x10^{15}$ (2000 pmoles) 2'-F pyrimidine modified RNA molecules. Resulting round 1 pools were mixed (at the transcription level) in equal portions for round 2. Random 2'-F pyrimidine modified RNA bound to hTGFß1 with an approximate K_D of ~10 nM. The rounds of the SELEX process were continued until no further improvement in K_D was observed. Figures 1A and 1B show binding curves of rounds 0, 14, 15L and 16L of the 40N pool (Fig. 1A) and rounds 0, 14, 15 and 17 of the 30N pool (Fig. 1B). The 40N pools showed the best affinity improvement followed by the 30N pool. The 20N pool showed no significant improvement after 12 rounds of SELEX. The RNA pools from the final rounds (round 16, 17 and 12 for the 40N, 30N and 20N, respectively) were reverse transcribed, PCR amplified and cloned as previously described (Pagratis *et al.* (1997) Nature Biotechnology 15:68-73). The 20N pool was cloned and sequenced as a control.

b) RNA sequences

The sequences of 64, 48, and 40 clones from the 40N, 30N and 20N final evolved pools, respectively, were determined and are summarized in Table 3 (SEQ ID NOS:6-143) in standard single letter code (Cornish-Bowden (1985) Nucleic Acid Res. 13:3021-3030).

Ligand designations in Table 3 include the size of the contributing random region followed by the ligand ID number. Ligands appearing more than once are designated with multiple ID numbers corresponding to their frequency. Ligands differing by one base are considered PCR derived variants of the same original molecule. Computer assisted global and local alignments suggest alignments and family assignments as shown in Table 4. There are 9 proposed families of which the first three include only 40N ligands. The remaining families contain clones derived from all three pools. However, it is clear from sequence lengths that cross contamination of the three pools had occurred. The possibility of cross contamination was minimized by electrophoretic size fractionation of RNA at each round, and PCR products prior to cloning.

Example 3. Binding Affinities of hTGFß1 Ligands

The dissociation constants of the hTGFß1 ligands were determined by nitrocellulose filter binding and are listed in Table 4. The majority of ligands bind hTGFß1 biphasically. Under conditions of protein excess, biphasic binding suggests that ligands can exist as two affinity species (presumably isoconformers) that are not in equilibrium, i.e. isoconformers that are kinetically trapped. The best identified ligands, 40-03 (SEQ ID NO:84) and 40-60 (SEQ ID NO:128) bind biphasically with the high and low affinity dissociation constant of ligand 40-03 at about 0.3 pM and 4.6 nM, respectively. There are observed variabilities in the K_D determinations for individual clones and random RNA, however, the high affinity species of ligands 40-03 and 40-60 always show about $>10^4$ better affinity than random RNA in any given experiment. A significant difference between random RNA and ligands 40-03 and 40-60 was also observed by BIAcore analysis. In the BIAcore analysis, biotinylated TGFß1 was coupled to a BIAcore chip and exposed to various concentrations of random RNA, ligand 40-03 and ligand 40-60. Also in this experiment the binding activities of ligands 40-03 and 40-60 were compared with the binding activity of an anti-TGFß1 polyclonal antibody (catalog No. AB-100-NA, R&D Systems, Minneapolis, MN). Figure 2 shows the ligand binding of the random RNA, ligands 40-03 and 40-60, and of the anti-TGFß1 antibody. From these Biacore data the determined dissociation rate constant (k off) for ligand 40-03, ligand 40-60 and anti-TGFß1 were about $2.7x10^{-4}$, $7.0x10^{-4}$ and $4.4x10^{-5}$, respectively. Therefore, ligands 40-03 and 40-60 show binding properties similar to the

control antibody with the off rate of 40-03 being about 6 fold faster than the off rate of the anti-TGFß1.

Example 4. Specificity of RNA Ligands to hTGFß1

The specificity of ligands 40-03 (SEQ ID NO:84) and 40-60 (SEQ ID NO:128) to TGFß1 was tested by comparing their dissociation constants with the closely related protein TGFß2 and the heparin binding human growth factors hVEGF and hKGF. The results summarized in Table 5 show that ligands 40-03 and 40-60 are specific for hTGFß1. Ligands 40-03 and 40-60 have binding affinities similar to random RNA to the other proteins tested. These ligands are four to five orders of magnitude more specific for TGFß1 than even closely related proteins such as TGFß2 and other heparin binding growth factors. Of particular interest is the ability of these TGFß1 ligands to discriminate between TGFß1 and TGFß2 since these two proteins share 72% identity and are interchangeable in most biological assays (Roberts and Sporn (1991), "The Transforming Growth Factor-ß's" in Peptide Growth Factors and Their Receptors, M. B. Sporn and A. B. Roberts, eds. (New York: Springer-Verlag)). Recently the solution three-dimensional structure of TGFß1 has been described and compared to the X-ray structure of TGFß2 (Hinck *et al.* (1996) Biochemistry 35:8517-8534). Based on this comparison there is only a slight structural difference between TGFß1 and TGFß2 with a maximum root mean square deviation of 1.9 Å (Hinck *et al.* (1996) Biochemistry 35:8517-8534). BIAcore technology was also utilized to compare the binding specificity of ligands 40-03 and 40-60 between TGFß1 and TGFß2. The analysis chip, loaded with either biotinylated 40-03, biotinylated 40-60, or biotinylated random RNA was exposed to various concentrations of TGFß1 or TGFß2 at 20 µL/min in HBSMC-HSA, and data was collected during the association phase (3 min) and the dissociation phase (3 min).

Figures 3A-3F show a typical nested series of sensorgrams with TGFß1 and TGFß2 binding to random RNA, ligand 40-03 and ligand 40-60. These BIAcore results show that when applied at high concentrations, TGFß1 binds random RNA (Fig. 3A), ligand 40-03 (Fig. 3B) and ligand 40-60 (Fig. 3C) equivalently in a nonspecific manner with fast on-rates and off-rates. This non-specific binding is low affinity and non-stoichiometric, since stoichiometric binding would result in about 500 RU's of TGFß1 bound to the RNA on the chip (see Example 1(f)). This non-specific binding represents the binding of random RNA

to TGFß1 also observed by nitrocellulose filter binding (see Example 2(a)). When applied at lower concentrations, (less than 50 nM) TGFß1 binds ligand 40-03 and 40-60 but not random RNA. The specificity of TGFß1 for ligands 40-03 and 40-60 is mainly due to slower off rates compared to random RNA. This represents a specific interaction which appears to be stoichiometric, since the binding curves at this concentration plateau at about 400 RU's and the dissociation rates are very slow. See, for example, the triangles in Figure 3B, in which the dissociation rate is almost flat.

TGFß2 behaves differently in the same experiment. TGFß2 shows no binding to random RNA (Fig. 3D) and some binding to ligand 40-03 (Fig. 3E) and ligand 40-60 (Fig. 3F). This difference in binding affinity to random RNA is consistent with the increased negative charge content of TGFß2 compared to TGFß1. The results in Figures 3D-3F clearly show that TGFß2 binds ligands 40-03 and 40-60 better than random RNA. However, the observed TGFß2 binding to ligand 40-03 and 40-60 is still different, and lower than the corresponding binding of TGFß1. It seems that TGFß2 binds ligand 40-03 and 40-60 with a very slow on and off rate suggesting induced fit. These results suggest that ligands 40-03 and 40-60 show cross-reactivity and bind to both TGFß1 and TGFß2 but with different affinities and kinetics.

Example 5. Inhibition of TGFß1 bioactivity

TGFß1 is a multifunctional growth factor (Roberts and Sporn (1991), "The Transforming Growth Factor-ß's" in Peptide Growth Factors and Their Receptors, M. B. Sporn and A. B. Roberts, eds. (New York: Springer-Verlag)). One of its activities is inhibition of proliferation of epithelial cells. For example, TGFß1 causes mink lung epithelial cells (MLEC) to cease replication, and it is manifested by reduction in ^{3}H-thymidine incorporation. The midpoint of this response of MLEC is about 0.3 pM.

RNA from round 11 and 14 of the 40N and 30N pools along with random RNA controls were tested for TGFß1 inhibitory activity using mink lung epithelial cells and measuring ^{3}H-thymidine incorporation in the presence of 2 pM hTGFß1. A significant hTGFß1 inhibitory activity was observed with these advanced pools and not with random RNA (Figures 4A and 4B). It appears that the 40N round 14 pool was neutralizing serum-derived TGFß1 in addition to the supplied TGFß1 since the amount of DNA synthesis at

high RNA concentrations is greater than that observed without exogenously added TGFß1 (Fig. 4A).

Using the same MLEC assay several individual ligands were screened for TGFß1 inhibitory activity. The results are summarized in Table 4 (Ki column). Several ligands were found that are good inhibitors of hTGFß1. Typical results are shown in Figures 5A-5D. It seems that the majority of good inhibitors belong in class 1 which contains only ligands from the 40N (Table 4, Ki column), and as expected, the best bioactivity correlated with binding activity.

TGFß1 proteins of various species are highly conserved proteins. The human and mouse or rat TGFß1 differ by a single amino acid. To determine the cross-species specificity, the ability of the TGFß1 ligands to inhibit the murine (m)TGFß1 bioactivity was tested. Since mTGFß1 is not commercially available, conditioned media from mouse cells was used. Several cell lines were screened for TGFß1 activity and it was found that 3T3 cells were the best source. Figure 6 shows the specificity of conditioned media used and the ability of ligand 40-03 and 40-60 to inhibit the bioactivity of such conditioned media. Inhibition profiles with a pan-specific antibody (monoclonal mouse anti-TGFß1/ TGFß2/TGFß3 antibody; Fig. 6, open triangles) and a TGFß2/TGFß3 specific antibody (Fig. 6, open circles) demonstrate that the ability of the 3T3 conditioned media to inhibit the growth of MLEC is mainly due to TGFß1. Figure 6 also clearly demonstrates that, as expected, ligands 40-03 and 40-60 can inhibit the bioactivity of the 3T3 CM, presumably due to mTGFß1.

Example 6. Effect of library random region length on the outcome of the SELEX

The above results suggest that size of the random region is important for the outcome of the SELEX process with TGFß1 in terms of obtaining bioactive ligands. These data are summarized in Table 6. It appears that the 30N pool contained ligands that bind TGFß1 with good affinities but these 30N ligands in general fail to inhibit the TGFß1 bioactivity. The 20N pool failed to yield any TGFß1 ligands. Only the 40N pool yielded ligands that bind TGFß1 and inhibit its bioactivity.

Example 7. Proposed secondary structure of bioactive TGFß1 Ligands

The predicted common secondary structures among those ligands that could inhibit TGFß1 bioactivity were investigated. These ligands appear to accommodate the proposed structure shown in Figure 7 which is a double pseudoknot. This structure is consistent with enzymatic digestion results obtained with three bioactive class 1 ligands. Such enzymatic digestion confirmed stem 1 and stem 2 while stem 3 was postulated on the basis of truncation results.

TABLE 1

Starting ssDNA templates

40N7:

5'GGGAGGACGATGCGG[-40N-]CAGACGACTCGCCCGA 3' SEQ ID NO: 1

30N7:

5'GGGAGGACGATGCGG[-30N-]CAGACGACTCGCCCGA 3' SEQ ID NO: 2

20N7:

5'GGGAGGACGATGCGG[-20N-]CAGACGACTCGCCCGA 3' SEQ ID NO: 3

SELEX PCR Primers:

5G7:

5'TAATACGACTCACTATAGGGAGGACGATGCGG 3' SEQ ID NO: 4

3G7:

5'TCGGGCGAGTCGTCTG 3' SEQ ID NO: 5

TABLE 2. TGFß1 SELEX conditions and results

Round	[P][1], nM	[R][2], nM	%B[3]	S/N[4]	PF[5]	PB[6]	Spin[7]	Bf. Wash[8]	U. Wash[9]
40N									
1A	100	5000	0.42	13	-	-	-	5	
1B	100	1000	0.60	30.7	-	-	-	5	
2	100	500	0.98	4.9	+	-	-	5	
3	100	500	3.40	2.6	+	-	-	10	
4	100	500	4.90	2.9	+	-	-	10	
5	33	167	2.50	1.9	+	-	+	10	5
6	33	167	ND	ND	+	-	+	10	55
7	11	56	1.00	8.0	+	+	+	10	55
8	11	56	0.40	5.0	+	+	+	10	55
9	3.3	16.5	ND	13.7	+	+	+	10	55
10	1.1	5.6	1.55	16.5	+	+	+	5	5
11	0.33	1.5	2.00	7.0	+	+	+	5	5
12*	0.03	0.15	1.31	8.0	+	+	+	5	5
13*	0.0033	0.016	0.33	2.4	+	+	+	5	5
14*	0.011	0.055	1.00	3.5	+	+	+	5	5
15L	0.033	0.0066	10.00	130.0	+	+	+	5	5
16L	0.033	0.0066	11.50	345	+	+	+	5	5
30N									
1A	140	7000	0.36	4.4	-	-	-	5	
1B	140	1400	1.80	20.9	-	-	-	5	
2	140	700	1.90	11.1	+	-	-	5	
3	140	700	4.60	4.4	+	-	-	10	
4	140	700	5.20	9.0	+	-	-	10	
5	5.0	25.6	1.50	4.3	+	-	+	10	5
6	11	55	0.70	2.6	+	-	+	10	55
7	3.3	16.5	0.26	1.7	+	+	+	10	55
8	3.3	16.5	0.10	2.0	+	+	+	10	55
9	3.3	16.5	ND	14.4	+	+	+	10	55
10	1.1	5.6	0.39	4.5	+	+	+	5	5
11	0.33	1.5	0.38	4.0	+	+	+	5	5
12*	0.03	.15	0.40	3.0	+	+	+	5	5
13*	0.03	.16	0.49	3.0	+	+	+	5	5
14	0.11	.55	0.90	10.0	+	+	+	5	5
15	0.033	0.165	0.50	6.7	+	+	+	5	5
16L	0.11	.022	1.8	25.7	+	+	+	5	5
17L	0.033	0.0066	1.5	13.6	+	+	+	5	5

Table 2 continued:

Round	[P][1], nM	[R][2], nM	%B[3]	S/N[4]	PF[5]	PB[6]	Spin[7]	Bf. Wash[8]	U. Wash[9]
20N									
1A	1000	50000	0.54	15.8	-	-	-	5	
1B	100	1000	1.70	39.5	-	-	-	5	
1C	1000	5000	3.80	51.0	-	-	-	5	
2	1000	5000	3.70	72.5	+	-	-	5	
3	1000	5000	5.90	122.0	+	-	-	10	
4	330	1670	1.70	17.4	+	-	-	10	
5	4.0	20.6	1.00	10.6	+	-	+	10	5
6	1.2	6.1	0.60	4.7	+	-	+	10	10
7	3.3	16.5	0.06	3.0	+	+	+	10	55
8	3.3	16.5	0.30	15	+	+	+	10	55
9	3.3	16.5	ND	6.6	+	+	+	10	55
10	3.3	16.5	0.31	16.5	+	+	+	5	5
11	1.1	5.6	0.19	4.0	+	+	+	5	5
12	1.1	5.6	1.2	13.0	+	+	+	5	5
13L	0.1	0.022	0.9	10.0	+	+	+	5	5

[1]Protein concentration in nanomolar
[2]RNA concentration in nanomolar
[3]Backround expressed as % of input
[4]Signal to noise
[5]Use of nitrocellulose prefiltered RNA
[6]Use of preblocked nitrocellulose with BSA
[7]Spinning of binding reactions before filtering through nitrocellulose
[8]Volume in ml of buffer wash
[9]Volume in ml of 0.5M urea wash
[10]L indicates RNA limiting SELEX conditions
[11]The RNA pool used was a mixture of 2-3 pools obtained from 3 fold serial dilutions of a binding reaction. Only the most stringent condition is shown.

TABLE 3. Sequence of individual TGFß1 RNA ligands. The sequences of the fixed regions (Table 1) are not shown.

		SEQ ID NO:
20-01	GUCUAUUUUUGCCUCCUCCC	6
20-02	AAUCCUUUCUUAAACCUCCC	7
20-03	UGUCUUUAGCUUAGGUUAUUCCUUCUGCCG	8
20-04	UGUCUUUAGCUUAGGUGAUUCCUUCUGCCG	9
20-05	UGUCUCUACCUUAGGUUGAUUCCUUCUACCG	10
20-06	UGAGUCUUGUUUUUUCGUC	11
20-07	UUGGCAUUGAAAGAGCUGGCAUACAUUCGC	12
20-08	UCCUUUCUAACAUUCCUCCC	13
20-09	GUCGUUGUUUUUCUCCUCCC	14
20-10	UGAGUCUUUCUUUUCGUCCC	15
20-11	GUCGUUUUUUUGGUCCUC	16
20-12	GUUUUUAUUAUUCGUUUGGC	17
20-14	GUCGAUCAUUUUUAGCCUCCC	18
20-17	UGAGUUGAUCUUUUCGUCCC	19
20-18	UGCCUUUAGCUUAGGCAUUGCCUUCUGUG	20
20-19	CAAAAUUUUUGGUCAAGCCGUCAUUGCCGC	21
20-21	GUCGUUCUUUUUUCCCUCCC	22
20-23	AAUUUUUGUGAAGACGUUUGCCGCUUUGCC	23
20-24	CGCAUCUUCUGUUUUCUCCC	24
20-25	GGAAUUUUUGGUAAAGCCGUAUGCCUCGC	25
20-26	UCAUCUCUGGGAGUUAAGAUCAUUUGGCCG	26
20-27	GCAGCCUCUGAUUUUCUCCC	27
20-28	GUCGUGAUUUUCGUUCUGCC	28
20-29	GUCGUAUUUUUUCCGCCUCCC	29
20-31	UCCUCAGCCUCUCACUUAUUAUCCUCCC	30
20-34	GUCUACUUGUUUUACCUCCC	31
20-35	CGAUUUUUUCGUCUUUUGGC	32
20-36	UGUCUAUAGCCUUGAUUAUAUCAUCUGCCG	33
20-37	CGAUUCCUCUUUUCACUCCC	34
20-38	UCCCAUUUUUCUCCUCUCCC	35
20-40	GUUAAUUUUUGUCCUCUGGC	36
20-41	UUUUUUUCUUUUUUCUUUUUUUCCG	37

Table 3 cont'd

		SEQ ID NO:
20-42	UCGUCUUUGUUUUUCUCCC	38
20-43	UGUCUAUAGCCUUGAUUACAUCAUCUGCCG	39
20-45	UGCCUUUAGCUUAGGCAUUGCCUUCUGCCG	40
20-46	UGUCUAUAGCUUGAUUUUUAAUUUCUGCCG	41
20-47	UUUUAUUUUCUUCGUCUGGC	42
20-48	GAUGAACCGAACCGAGGUUAAGGUGCCAGAGUAGACGCUCAU	43
20-49	UCGUCUAUUUUUUCCCUCCC	44
20-50	CUUUCGUCUGUUUUCCUGCC	45
30-01,07,18,23	UGUCUUUAGCCUAGGUGAUUCCUUCUGCCG	46
30-02	CCUUGUUUUCUUUUUUCUUUUUUCACCCC	47
30-03	UGUCUUUAGCCCAGGUGAUUCCUUCUGCCG	48
30-04	UUAACCGUAAAGACGGCAUGAUGUAGUCCG	49
30-05	UUUUUUUAGCUUAGGUGAUUCCUUCNNCCU	50
30-06	UGCCUUUAGCUUAGGCUUUGCCUUCUGCCG	51
30-08	CGGAAUUUUUGUUGAGCCGUAUGCCGC	52
30-09,42	UGCCUUUAGCUUAGGUGAUUCCUUCUGCCG	53
30-10	UGUCUUUAGCCUAGGUGAUUCCUUCUGCCG	54
30-12,24,21,40,41	UGUCUAUAGCCUGAUUUUUAAUCUCUGCCG	55
30-15	UUGACCGUUAAGACGGCAUGAUGUGGUCCG	56
30-16,27,38,46	UGCCUUUAGCUUAGGCAUUGCCUUCUGCCG	57
30-17	UGCCUUUAGCUUAGGCUUUGCCUUCUGCCG	58
30-19	UUAACCNUAAAUACGGCUUGANUUCUUCCG	59
30-20	UGCCUUUAGCUUAGGCAUUGCCUUCUGCCG	60
30-22	UUAACCGUAAAGACGGCAUGAUGUUUUCCG	61
30-25	UUGGCAUUGAAAGAGGCGUCAUAUGUUCGC	62
30-26	CCUUUCUUUCUUUUUAUUUUCUUCCCCUCCC	63
30-28	UGCCUUUAGCCUAGACCUUGUCUUCUGCCG	64
30-29	UGUCUUUAGCCUAGGUGAUUCCUUCUGCCG	65
30-30	UGUCUUUAGCCUAGGUGAUUCCUUCUGCCG	66
30-31	ACCGGUAAGGGCACUGCAGGAACACAAUCCCCUAUGCGAC	67
30-32	GGAAUUUUUGGUAAAGCCGUAUGCCUCGC	68
30-33	UGGCAUUGAAAGAGAUCGCAUACCUUCGC	69
30-34	UGUCUAUAGCCUUGAUUACAUCAUCUGCCU	70
30-35	UGUCUUUAGCCUAGGUGAUUCCUUCUGCCU	71

Table 3 cont'd

		SEQ ID NO:
30-14	UGCCUUUAGCUUAUGCAUUGCCUUCUGCCG	72
30-36	UGCCUUUAGCUUAGGCAUUCGCCUUCUGCCG	73
30-37	UGUCUUUGGCCUAGGUGAUUCCUUCUGCCG	74
30-39	UGUCUUUAGCUUAGGUGAUUCCUUCUGCCG	75
30-43	UGUCUUUAGCCUAGGUGAUUCCUUCUGCCG	76
30-44	UGCCUUUAGCUUAGGCAUUGCCUUGCCG	77
30-45	GGUCUUUUAUUUUUUGUUUUUCUCUGUGCCC	78
30-47	UUAACCGUAAAGACAGCAUGAUGUAGUCUG	79
30-48	UUUUUUUCUUUUCCUUCCUUUUCUUACCG	80
30-49	UUAACCGUAAAGACGGCAUGAUGUUGUCCG	81
30-50	GGAAUUUUUGGUAAAGCCGUAUGCCUCGC	82
40-02	GCCAAGGUUACGCCGUCGGACCCUGCUGCCAACAUCCUCCC	83
40-03	GGGUUAUUGGGCGUCAACAUCCCCGAUUCUUUUCACGUC	84
40-04	AUGCCUUUUGCCUUCAGGGUGUAAUUCCUUGAUCUGUCCG	85
40-05	AACAAGGUUACGCCGUCGGACCCUGCUGCCAACAUCCUCCC	86
40-06	UUAGGGGCGUCAACACCGCUAUCAUAAUUUUCGCCUUCCC	87
40-08	CGCAAGGUUACGCCGUCGGACCCUGCUGCCAACAUCCUCC	88
40-11	UGCCUUUAGUCUGAAUCUUCUACCAUGAUUCUCUGCCG	89
40-12	GACCCUUGUCUGCGAUUCAACUCGUAGGUUUUCUCACGUG	90
40-13	AGCAAGGUUACGAGGUCGGACCCUGCUGCCAACAUCCUCCC	91
40-14	CAUUAUGGCGUCAACAUGCCGGUUUUCGAUUCUCAUUGUC	92
40-15	CUCUAACUUCUUUUUCGCCUGUGUGUUUUCUUUUUGCUG	93
40-16	UUAGGGGCGUCAACACCGCUAUUACAUCUUUCGCCUCCC	94
40-17	GGUCGUUUUGUUUUUGUUUUUUGUAGCCCGGUCAUCCC	95
40-19	UUAGCGCGAGUUCAACACCGCAUGUGAUUCUUUCGCCUCC	96
40-20	UACAAGGUUACGCCGUCGGACCCUGCUGCCAACAUCCUCCC	97
40-21,34	GACCCUUGUCUGCGAUUCAACUCGUAGGUCUUCUCACGUG	98
40-22,35	UUAGGGGCGUCAACACCGCUAUUACAAUUUUCGCUUCC	99
40-23	UUAGGGGCGUCAACACCGCUAUUACAAUCUUCGCUUCC	100
40-24	UUAUGGGCGUCAACACCGCUAUUACAACUUUCGCUUUCC	101
40-25	UGUCGAUCGUUUGCUGUUUGAUUUCUUUUGUCCCUCCCGUG	102
40-26	UUAGGGGCGUCAACAUCGCUAUUACAAUCUUCGCCUUCC	103
40-28	UUAGGGGCGUCAACACCGCUAUUACAACUUUCGCCUCAC	104
40-29	GACCCUUUUCUGCGAUUCAACUCGUACGUCUUCUCACGUG	105

Table 3 cont'd

		SEQ ID NO:
40-31	UUAAGGGCGUCAACACCGCUAUUACAACUUUCGCUUCC	106
40-32	UUAUGGGCGUCAACACCGCUAUUACAACUUUCGCCUC	107
40-33	AGCAAGGUUACGCCGUCGGACCCUGCUGCCAACAUCCUCCC	108
40-36	GUCAAGGUUACGCCGUCGGACCCUACUGCCCC	109
40-37	CUCCUAUAUUCAUGUUAUUGUUUUUUUCUUCCAGCUUGCCC	110
40-38	AGAUAAUUAUCAGCGGUGGACGGGGUGCCGGUACUGCCGC	111
40-39	UGCCUUUAGCCUAAGUUGAUCUAUUCAGCUUUCUGCCG	112
40-40	CCCAAGGUUACGCCGUCGGACCCUACUGCCAACUUCCUCCC	113
40-41	UGCCUUUAGCCUGAGUAUACUGAUGUAUAUUCUCUGCCG	114
40-42	UAGCGCGAGUUCAACACCGCAUGUGACUCUUUCGCCUCC	115
40-43	AUCCUUUUUUUAGCUUUUUUCUUUUUCCUGCCCCACUUCCC	116
40-44	UGCAAGGUUACGCCGUCGGACCCUGCUGCCAACAUCCUCCC	117
40-45	GGGCUUUUCCUUUAGUACUUUUUUGUUUCGCUCCCCCC	118
40-51	UGCCUUUAGUCUGAAUCUUACCAUGCGAUUUUCUGCCG	119
40-52	AACAAGGUUACUCCGUCGGACCCUGCUGCCAACAUCCUCCC	120
40-53	GACUCUUGUCUGCGAUUCAACUCGUAGGUCUUCUCACGUG	121
40-54	UUAGGGGCGUCAACACCGCUAUCAUAACUUUCGCUUCCC	122
40-55	UUAGGGGCGUCAACACCGCUAUUCAACCUUCGCUUCCC	123
40-56	UUAGGGCGUCAACACCGCUAUUACAACUUUCGCCUCCC	124
40-57	GGUGUCGUCUUUCAACCCCU	125
40-58	UUAUGGGCGUCAACACCGCUAUUACAACUUUCGCCUCCC	126
40-59	CCCAAGGUUACGCCGUCGGACCCUGCUGCAAACAUCCUCCC	127
40-60	UUAUGGGCGUCAACACCGCUAUUACAGUUUUCGCCUCCCC	128
40-61,76	UUAGGGGCGUCAACACCGCUAUUACAAUCUUCGCUUUCC	129
40-62	GCCAAGGUUACGCCGUCGGACCCUGCUGCCAACAUCUUCCC	130
40-64	UUAGGGGCGUCAACACCGCUAUUACAAUCUUCGUCUUCC	131
40-65	GUCAAGUUUACGCCGUCGGACCCUGCUGCCAACAUCCUCCC	132
40-66	UUCAAGGUUACGCCGUCGGACCCUGCUGCCAACAUCCUCCC	133
40-67	CUCAAGGUUACGCCGUCGGACCCUGCUGCCAACAUCCUCCC	134
40-68	UUAGGGGCUUCAACACCGCUAUUACAUUCUUCGCCUCCC	135
40-69	CACAAAGUUACGCCGUAGGACCCUGCUGCCAACAUCCUCCC	136
40-70	GGAUGGUCAGUUUCGGUUUUUCAUAUGUUUAUUUUCCCCCC	137
40-71	UAUUGACUUUUGUUUCUUUUUCUUUGCCUGGUCCC	138

Table 3 cont'd

		SEQ ID NO:
40-72	UUAGGGGCGUCAACACCGCUAUUACAACUUUCGCUUCCC	139
40-73	CUUCUUUUUCUUCUUUUCUUUAUGUCUUCUUCAUGCCG	140
40-75	GACCNUUGUNUGCGAUUCAACUCGUAGGUCUUCUCACGUG	141
40-77	UUAUGGGCGUCAACACCGCUAUUACAACUUUCGCCCCC	142
40-79	UUAUGGGUGUCAACACCGCUAUUACAACUUUCGCCUCCC	143

TABLE 4. Proposed alignment and observed affinity and bioactivity of TGFß1 ligands. The sequences of the fixed region (Table 1) are not shown.

Class 1

			Kd1 (nM)	Kd2 (pM)	P1(%)	P2(%)	P2/P1(%)	Ki, nM
40-03	GGGUUA UUGGGCGUCAACAUCCCCGAU UCUUUUCA CGUC		4.6±1.1	0.3±0.08	60.3	24.4	40.5	0.4
40-06	UUA GGGGCGUCAACACCGCU AU CAUAAUUUU CGCCUUCCC		3.7±0.6	1.6±0.6	60.0	10.0	16.7	
40-14	CAUUA UGGCGUCAACAU GCCGGUUUUCGAUUCUCAUUGUC		5.7±1.4	0.4±0.2	62.9	12.1	19.2	3.9
40-16	UUA GGGGCGUCAACACCGCU AU UACA UCUUU CGCCUCCC		1.7±0.6	0.06±0.04	48.5	32.8	67.6	0.6
40-19	UUAGCGCGAGUUCAACACCGC AU GUGAUUCUUU CGCCUCC		4.2±2.2	3.7±2.7	68.0	21.5	31.6	28.0
40-22, 35	UUA GGGGCGUCAACACCGCU AU UACAAUUUU CGCUUCC		13.9±4.3	17.6±4.5	93.8	13.3	14.2	
40-23	UUA GGGGCGUCAACACCGCU AU UACAAUCUU CGCUUCC		14.2±4.5	15.6±4.0	58.2	13.1	22.5	0.75
40-24	UUA UGGGCGUCAACACCGCU AU UACAACUUU CGCUUUCC		12.7±4.7	27.5±10.2	100	12.4	12.4	
40-26	UUA GGGGCGUCAACAUCGCU AU UACAAUCUU CGCCUUCC		7.7±1.6	40.8±23.3	73.7	8.4	11.4	
40-28	UUA GGGGCGUCAACACCGCU AU UACAACUUU CGCCUCAC		12.3±2.4	81.6±47.2	93.4	4.8	5.1	
40-31	UUA AGGGCGUCAACACCGCU AU UACAACUUU CGCUUCC		8.4±2.8	0.7±0.4	34.4	9.3	27.0	2.56
40-32	UUA UGGGCGUCAACACCGCU AU UACAACUUU CGCCUC		14.0±6.4	5.0±3.3	51.1	7.7	15.0	
40-42	UAGCGCGAGUUCAACACCGC AU GUGACUCUUU CGCCUCC		11.4±1.9	0.09	37.9	6.2	16.3	
40-54	UUA GGGGCGUCAACACCGCU AU CAUAACUUU CGCUUCCC		8.5±1.8	6.75±9	46.7	25.9	55.5	
40-55	UUA GGGGCGUCAACACCGCU AU U CAACCUU CGCUUCCC		8.0±2.5	0.1±0.06	64.9	24.8	38.2	2.9
40-56	UUA GGGCGUCAACACCGCU AU UACAACUUU CGCCUCCC		4.2±1.3	0.2	41.0	12.3	30.0	7.4
40-58	UUA UGGGCGUCAACACCGCU AU UACAACUUU CGCCUCCC		4.4±1.6	5.3±2.2	35.1	24.6	70.0	0.9
40-60	UUA UGGGCGUCAACACCGCU AU UACAGUUUU CGCCUCCCC		3.8±1.5	1.9±0.8	32.5	25.5	78.5	0.8
40-61, 76	UUA GGGGCGUCAACACCGCU AU UACAAUCUU CGCUUUCC		13.5±4.2	1.0±0.8	56.5	4.9	8.6	
40-64	UUA GGGGCGUCAACACCGCU AU UACAAUCUU CGUCUUCC		5.6±2.0	3.1±1.8	37.0	15.3	41.3	
40-68	UUA GGGGCUUCAACACCGCU AU UACAUUCUU CGCCUCCC		20.7±3.2	0.4±0.3	62.9	1.9	3.0	
40-72	UUA GGGGCGUCAACACCGCU AU UACAACUUU CGCUUCCC		1.4±0.8	0.07±0.05	23.8	42.4	?	
40-77	UUA UGGGCGUCAACACCGCU AU UACAACUUU CGCCCCC		3.7±1.5	1.73±1.43	27.0	16.3	60.4	
40-79	UUA UGGGUGUCAACACCGCU AU UACAACUUU CGCCUCCC		0.6±0.1	0.5±0.4	18.5	12.23	66	

Table 4 cont'd

Class 2

		Kd1 (nM)	Kd2 (pM)	P1(%)	P2(%)	P2/P1(%)	Ki, nM
40-02	GCCAAGGUUACGCCGUCGGACCCUGCUGCCAACAUCCUCCC	14.8±1.4		100			
40-05	AACAAGGUUACGCCGUCGGACCCUGCUGCCAACAUCCUCCC	12.6±		100			
40-08	CGCAAGGUUACGCCGUCGGACCCUGCUGCCAACAUCCUCC	15.4±1.6		82			
40-13	AGCAAGGUUACGAGGUCGGACCCUGCUGCCAACAUCCUCCC	15.4±9.6		100			
40-20	UACAAGGUUACGCCGUCGGACCCUGCUGCCAACAUCCUCCC	11.2±7.8		100			>1300
40-33	AGCAAGGUUACGCCGUCGGACCCUGCUGCCAACAUCCUCCC	26.6±14		100			
40-36	GUCAAGGUUACGCCGUCGGACCCUACUGCCCC	41.7±9.5		99			
40-40	CCCAAGGUUACGCCGUCGGACCCUACUGCCAACUUCCUCCC	18±2.8		91.3			
40-44	UGCAAGGUUACGCCGUCGGACCCUGCUGCCAACAUCCUCCC	11.1±0.8		60.44			
40-52	AACAAGGUUACUCCGUCGGACCCUGCUGCCAACAUCCUCCC						
40-59	CCCAAGGUUACGCCGUCGGACCCUGCUGCAAACAUCCUCCC						
40-62	GCCAAGGUUACGCCGUCGGACCCUGCUGCCAACAUCUUCCC						
40-65	GUCAAGUUUACGCCGUCGGACCCUGCUGCCAACAUCCUCCC						

		Kd1 (nM)	Kd2 (pM)	P1(%)	P2(%)	P2/P1(%)	Ki, nM
40-66	UUCAAGGUUACGCCGUCGGACCCUGCUGCCAACAUCCUCCC						
40-67	CUCAAGGUUACGCCGUCGGACCCUGCUGCCAACAUCCUCCC						
40-69	CACAAAGUUACGCCGUAGGACCCUGCUGCCAACAUCCUCCC						

Class 3

		Kd1 (nM)	Kd2 (pM)	P1(%)	P2(%)	P2/P1(%)	Ki, nM
40-12	GACCCUUGUCUGCGAUUCAACUCGUAGGUUUUCUCACGUG	5.5±0.7	0.7±0.2	64.7	10.0	15.5	6.88
40-21, 34	GACCCUUGUCUGCGAUUCAACUCGUAGGUCUUCUCACGUG	10.1±3.5	6.5±4.0	100	9.3	9.3	1.68
40-29	GACCCUUUUCUGCGAUUCAACUCGUACGUCUUCUCACGUG	10.9±5.5		100			
40-53	GACUCUUGUCUGCGAUUCAACUCGUAGGUCUUCUCACGUG						>1300
40-75	GACCNUUGUNUGCGAUUCAACUCGUAGGUCUUCUCACGUG						2.25

Table 4 cont'd
Class 4

		Kd1 (nM)	Kd2 (pM)	P1 (%)	P2 (%)	P2/P1 (%)	Ki, nM
20-03	UGUCUUUAGCUUAGG						
20-04	UGUCUUUAGCUUAGG						
20-05	UGUCUCUACCUUAGG						
20-18	UGCCUUUAGCUUAGG						
20-36	UGUCUAUAGCCUUGA						
20-43	UGUCUAUAGCCUUGA						
20-45	UGCCUUUAGCUUAGG						
20-46	UGUCUAUAGCUUGAU						
30-01,07, 18,23	UGUCUUUAGCCUAGG						
30-03	UGUCUUUAGCCCAGG						
30-05	UUUUUUUAGCUUAGG						
30-06	UGCCUUUAGCUUAGG						
30-09,42	UGCCUUUAGCUUAGG						
30-10	UGUCUUUAGCCUAGG						
30-12,24, 21,40,41	UGUCUAUAGCCUGAU						
30-14	UGCCUUUAGCUUAUG						
30-16,27, 38,46	UGCCUUUAGCUUAGG						
30-17	UGCCUUUAGCUUAGG						
30-20	UGCCUUUAGCUUAGG						
30-28	UGCCUUUAGCCUAGA						
30-29	UGUCUUUAGCCUAGG						
30-30	UGUCUUUAGCCUAGG						
30-34	UGUCUAUAGCCUUGA						
30-35	UGUCUUUAGCCUAGG						
30-36	UGCCUUUAGCUUAGG						
30-37	UGUCUUUGGCCUAGG						
30-39	UGUCUUUAGCUUAGG						
30-43	UGUCUUUAGCCUAGG						
	UUAUUCCU UCUGCCG	0.11±0.1	0.3±1.7	9.3	13.4		
	UGAUUCCU UCUGCCG	0.2		8.0			
	UUGAUUCCU UCUACCG	0.11±0.1		10.49			
	CAUUGCC UUCUGUG						
	UUAUAUCA UCUGCCG						
	UUACAUCA UCUGCCG						
	CAUUGCCU UCUGCCG						
	UUUUAAUU UCUGCCG						
	UGAUUCCU UCUGCCG	3.4		63			
	UGAUUCCU UCUGCCG	58.2	262	~100	1.2		
	UGAUUCCU UCNNCCU	100					
	CUUUGCCU UCUGCCG	56.5	74.1	~100	2.4		
	UGAUUCCU UCUGCCG	5.35±0.9		22.7			
	UGAUUCCU UCUGCCG	4.05±1.5	75.5	28.6	17.9	62.6	>300
	UUUUAAUC UCUGCCG	2.82±1.7	60.8	24.8	23.2	93.5	
	CAUUGCCU UCUGCCG						
	CAUUGCCU UCUGCCG	71.8	211±90	99.8	4.3	~4	
	CUUUGCCU UCUGCCG	67	205±93	99.9	4.3	~4	
	CAUUGCCU UCUGCCG	2.57±0.3	0.7	22.9	6.1	26.6	
	CCUUGUCU UCUGCCG	0.78±.07	0.4±0.2	17.9	8.4	46.9	>130
	UGAUUCCU UCUGCCG						
	UGAUUCCU UCUGCCG						
	UUACAUCA UCUGCCU	9.5±1.6		68.5			
	UGAUUCCU UCUGCCU	20.7±13.8	224±123	90.3	8.8	9.7	
	CAUUCGCCUUCUGCCG	3.9±1.1		46.6			
	UGAUUCCU UCUGCCG	2.65±0.7		46.6			
	UGAUUCCU UCUGCCG	6.02±1.5	32.5±16.2	48.5	13.3	27.4	
	UGAUUCCU UCUGCCG	50.5		100			

Table 4 cont'd

		Kd1 (nM)	Kd2 (pM)	P1 (%)	P2 (%)	P2/P1 (%)	Ki, nM
30-44	UGCCUUUAGCUUAGG CAUUGC CUUGCCG						
40-04	AUGCCUUUUGCCUUCAGGGUGU AAUUCCUUGAUC UGUCCG	5.0±0.6	1.4±1.2	56.7	4.2	7.4	
40-11	UGCCUUUAGUC UGAAUCUUCUACCA UGAUUC UCUGCCG	4.6±0.7	4.9±3.1	68.9	8.0	11.6	
40-39	UGCCUUUAGCC UAAGUUG AUCUAUUCAGCUU UCUGCCG	11.5±2.0	8.78±6.4	64.2	3.6	5.6	
40-41	UGCCUUUAGCC UGAGUAU ACUGAUGUAUAUUC UCUGCCG	3.8±0.9	1.4±1.1	30.8	11.1	36.0	>130
40-51	UGCCUUUAGUC UGAAUCUU ACCAUGCGAUUU UCUGCCG						

Class 5

		Kd1 (nM)	Kd2 (pM)	P1(%)	P2(%)	P2/P1(%)	Ki, nM
20-26	UCAUCUCUGGGAGUUAAGAUCAUUUGGCCG						
30-04	UUAACCGUAAAGACGGCAUGAUGUAGUCCG	5.03±0.8		33.2			
30-15	UUGACCGUUAAGACGGCAUGAUGUGGUCCG	55.3		95.1			
30-19	UUAACCNUAAAUACGGCUUGANUUCUUCCG	5.7±1.9	346	28.8	13.6	47.2	53-52
30-22	UUAACCGUAAAGACGGCAUGAUGUUUUCCG	2.47		32.2			
30-47	UUAACCGUAAAGACAGCAUGAUGUAGUCUG						
30-49	UUAACCGUAAAGACGGCAUGAUGUUGUCCG						

Class 6

		Kd1 (nM)	Kd2 (pM)	P1(%)	P2(%)	P2/P1(%)	Ki, nM
20-19	CAAAAUUUUUGGUCAAGCCGUCAUUGCCGC						
20-23	AA UUUUUGUGAAGACGUU UGCCGCUUUGCC						
20-25	GGAAUUUUUGGUAAAGCCG UA UGCCUCGC						
30-08	CGGAAUUUUU GUUGAGCCG UA UGCCGC	10.2±2.9		33.8			
30-32	GGAAUUUUUGGUAAAGCCG UA UGCCUCGC						
30-50	GGAAUUUUUGGUAAAGCCG UA UGCCUCGC						

Class 7

		Kd1 (nM)	Kd2 (pM)	P1(%)	P2(%)	P2/P1(%)	Ki, nM
20-07	UUGGCAUUGAAAGAGCUGGCAUACAUUCGC						
30-25	UUGGCAUUGAAAGAGGCGUCAUAUGUUCGC	23±6.4	1.14±.6	79	2.7	3.4	
30-33	UGGCAUUGAAAGAGAUCGCAUACCUUCGC						

Table 4 cont'd

Class 8

		Kd1 (nM)	Kd2 (pM)	P1(%)	P2(%)	P2/P1(%)	Ki, nM
20-48	GAUGAACCGAACCGAGGUUAAGGUGCCAGAGUAGACGCUCAU						
30-31	ACCGGUAAGGGCACUGCAGGAACACAAUCCCCUAUGCGAC	100					
40-38	AGAUAAUUAUCAGCGGUGGACGGGGUGCCGGUACUGCCGC	21.8±5.3		92.7			

Class 9

		Kd1 (nM)	Kd2 (pM)	P1 (%)	P2 (%)	P2/P1 (%)	BCG (%)	Ki, nM
20-01	GUCUAUUUUU GCCUCCUCCC							
20-02	AAUCCUUUCUUAAA CCUCCC	0.6		5			0.6	
20-06	UGAGUCUUGUUUUUU CGUC	0.3		5			0.5	
20-08	UCCUUUCUAACAUU CCUCCC							
20-09	GUCGUUGUUUUU CUCCUCCC							
20-10	UGAGUCUUUCUUUU CGUCCC							
20-11	GUCGUUUUUUU GGUCCUC							
20-12	GUUUUUAUUAUUCGUUUGGC							
20-14	GUCGAUCAUUUUU AGCCUCCC							
20-17	UGAGUUGAUCUUUU CGUCCC							
20-21	GUCGUUCUUUUUU CCCUCCC							
20-24	CGCAUCUUCUGUUUUCU CCC							
20-27	GCAGCCUCUGAUUUUCU CCC							
20-28	GUCGUGAUUUU CGUUCUGCC							
20-29	GUCGUAUUUUUU CCGCCUCCC							
20-31	UCCUCAGCCUCUCACUUAUUAUCCUCCC							
20-34	GUCUACUUGUUUU ACCUCCC							
20-35	CGAUUUUUUCGUCUUUUG GC							
20-37	CGAUUCCUCUUUUC ACUCCC							
20-38	UCCCAUUUUU CUCCUCUCCC							
20-40	GUUAAUUUUUGUCCUCUGGC							
20-41	UUUUUUUCUUUUUUCUUUUUUU CCG							
20-42	UCGUCUUUGUUUUU CUCCC							
20-47	UUUUAUUUUCUU CGUCUGGC							

Table 4 cont'd

		Kd1 (nM)	Kd2 (pM)	P1 (%)	P2 (%)	P2/P1 (%)	BCG (%)	Ki, nM
20-49	UCGUCUAUUUUUU CCCUCCC							
20-50	CUUUCGUCUGUUUU CCUGCC							
30-02	CCUUGUUUUCUUUUUUCUUUUUUCACCCC	5.3±2.8	0.8±0.9	27.1	12.1	44.6	22.0	>1300
30-26	CCUUUCUUUCUUUUUAUUUUCUU CCCCUCCC						27.0	
30-45	GGUCUUUUAUUUUUUGUUUUUCU CUGUGCCC							
30-48	UUUUUUUCUUUUCCUUCCUUUUCUUACCG							
40-15	CUCUAACUUCUUUUUCGCCUGUGUGUUUUCUUUUU GCUG						31.6	
40-17	GGUCGUUUUGUUUUUGUUUUUUGUAGCCCGGUCAUCCC						35.1	
40-25	UGUCGAUCGUUUGCUGUUUGAUUUCUUUU GUCCCUCCCGUG						51.9	
40-37	CUCCUAUAUUCAUGUUAUUGUUUUUUUCUU CCAGCUUGCCC						29.6	
40-43	AUCCUUUUUUUAGCUUUUUUCUUUUU CCUGCCCCACUUCCC	2.2±1.7		12			13.2	
40-45	GGGCUUUUCCUUUAGUACUUUUUUGUUU CGCUCCCCCC						10.7	
40-57	GGUGUCGUCUUUC AACCCCU							
40-70	GGAUGGUCAGUUUCGGUUUUU CAUAUGUUUAUUUUCCCCCC							
40-71	UAUUGACUUUUGUUUCUUUUUCUUUGCCUGGUCCC							
40-73	CUUCUUUUUCUUCUUUUCUUUAUGUCUUCUU CAUGCCG							

Table 4 continued

Kd1 = Dissociation rate constant in nanomolar of the low affinity component of biphasic binding curves or dissociation rate constant in nanomolar of monophasic binding curves

Kd2 = Dissociation rate constant in picomolar of the high affinity component of biphasic binding curves

P1 = Plateau values in % of monophasic curves or of the low affinity component of biphasic curves

P2 = Plateau values in % of the high affinity component of biphasic curves

P2/P1 = Fraction in % of the high affinity component of biphasic curves

Ki = Inhibition constant in nanomolar obtained from the MLEC assay

BCG = Nitrocellulose binding background expressed as % of input

TABLE 5. Binding Specificity of TGFß1 Ligands 40-03 and 40-60

Target	$K_D^{Target} / K_D^{hTGFß1}$ 40-03	$K_D^{Target} / K_D^{hTGFß1}$ 40-60
hTGFß1	1	1
hTGFß2	>340,000	>340,000
hKGF	>34,000	>34,000
hVEGF	>340,000	>340,000

When applicable, the high affinity component of biphasic binding was used.

TABLE 6. Results of TGFß1 SELEX with random regions of 20 30 and 40N expressed by the distribution of ligands in the different classes and the binding and inhibitory activity of these classes

	SELEX Pools			Affinities			
	40N	30N	20N	Biph[1].	K_D~pM[2]	K_i[3]	
Total clones	64	48	40				
Unique clones	61	37	40				
Class 1	39.3%			+	+	+++	
Class 2	26.2%			-	-	-	
Class 3	8.2%			+	+	++	
Class 4	8.2%	56.7%	20.0%[4]	+	+	±	
Class 5		16.2%	2.5%[4]	+	+	+	
Class 6		8.1%	7.5%[4]	-	-	ND	
Class 7		5.4%	2.5%[4]	±	-	ND	
Class 8	1.6%	2.7%	2.5%[4]	-	-	ND	
Class 9	16.4%	10.8%	65.0%		NC[5]	-	
Length of 40	59 (96.7%)	1 (2.7%)			1 (2.5%)		
Length of 30	1 (1.6%)	36 (97.3%)			15 (37.5%)		
Length of 20	1 (1.6%)		24 (60.0%)				

[1]Biphasic binding is shown by plus (+), monophasic by minus (-), and unclear results by plus/minus (±)
[2]Low pmolar K_D values are shown by plus (+) and K_D values similar to random RNA are shown by minus (-)
[3]High, intermediate, low, and possible bioactivity is shown by 3 pluses (+++), two pluses (++), one plus (+) or plus/minus (±), respectively
[4]longer than 20N
[5]nitrocellulose binders

WE CLAIM:

1. A purified and isolated non-naturally occurring RNA ligand to TGFβ1 wherein said ligand is selected from the group consisting of the sequences set forth in Table 3 (SEQ ID NOS: 6-143).

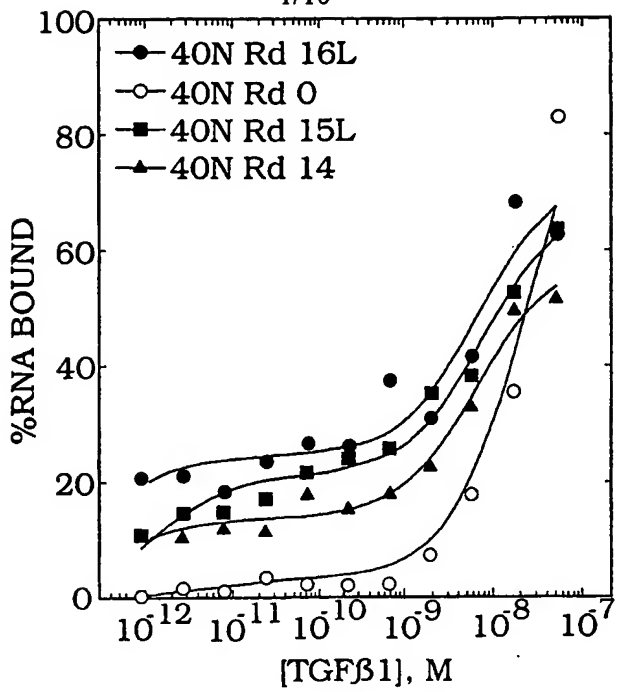


Figure 1A

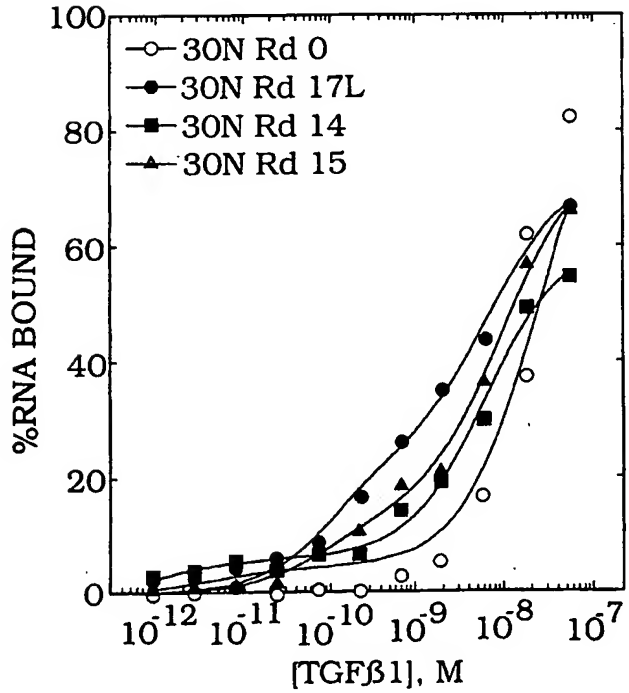
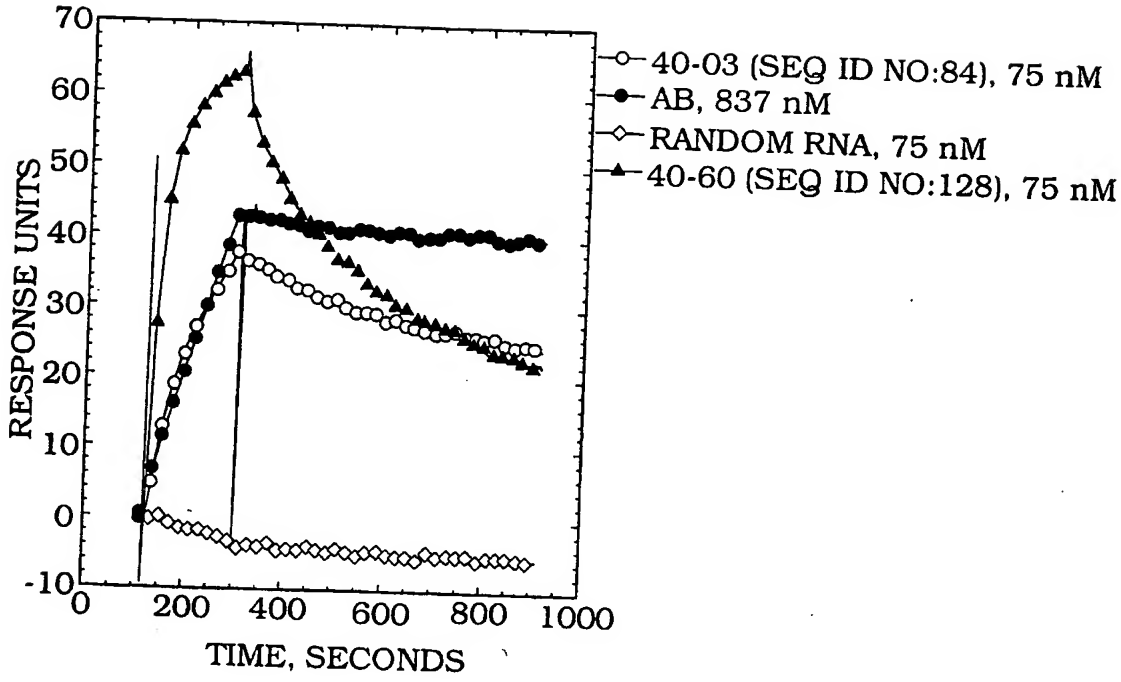


Figure 1B


70
60
50
40
30
20
10
0
-10
RESPONSE UNITS
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200
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600
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TIME, SECONDS
40-03 (SEQ ID NO:84), 75 nM
AB, 837 nM
RANDOM RNA, 75 nM
40-60 (SEQ ID NO:128), 75 nM

Figure 2

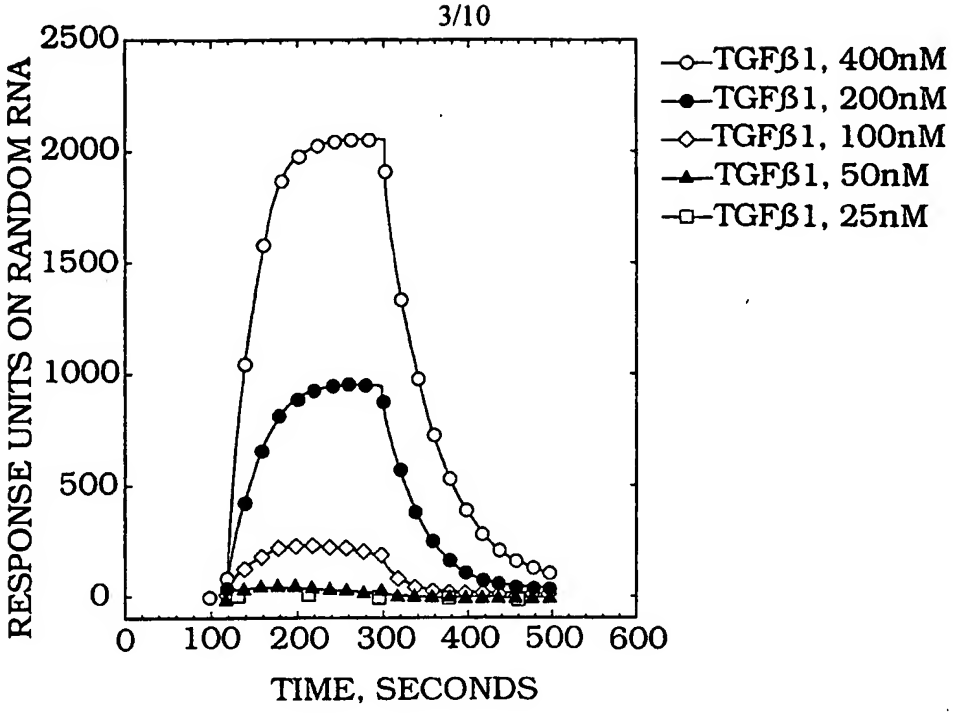

Figure 3A

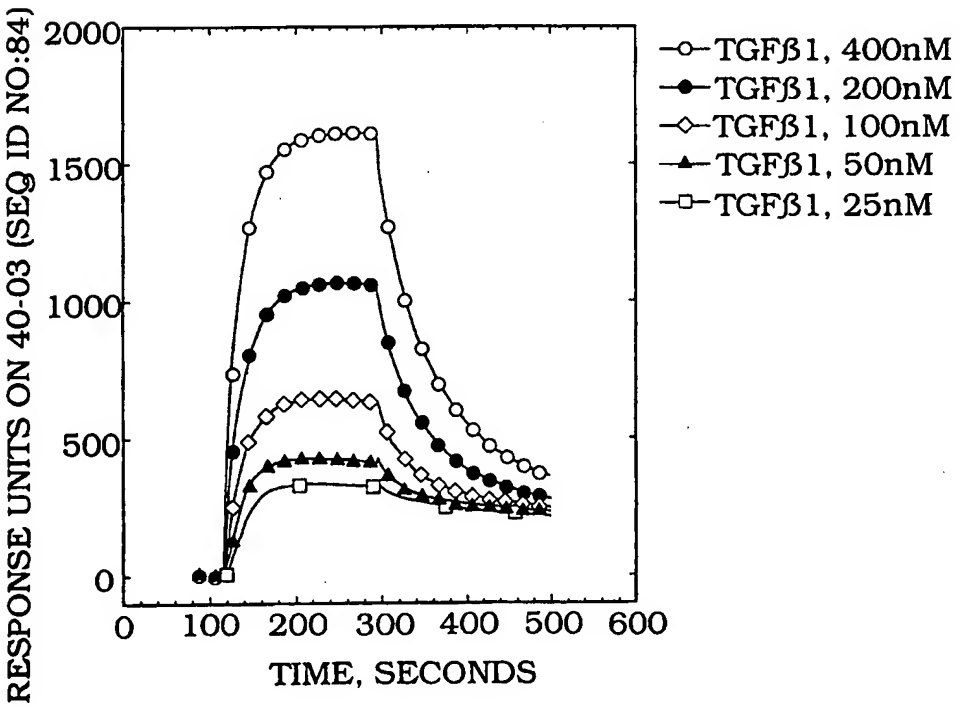

Figure 3B

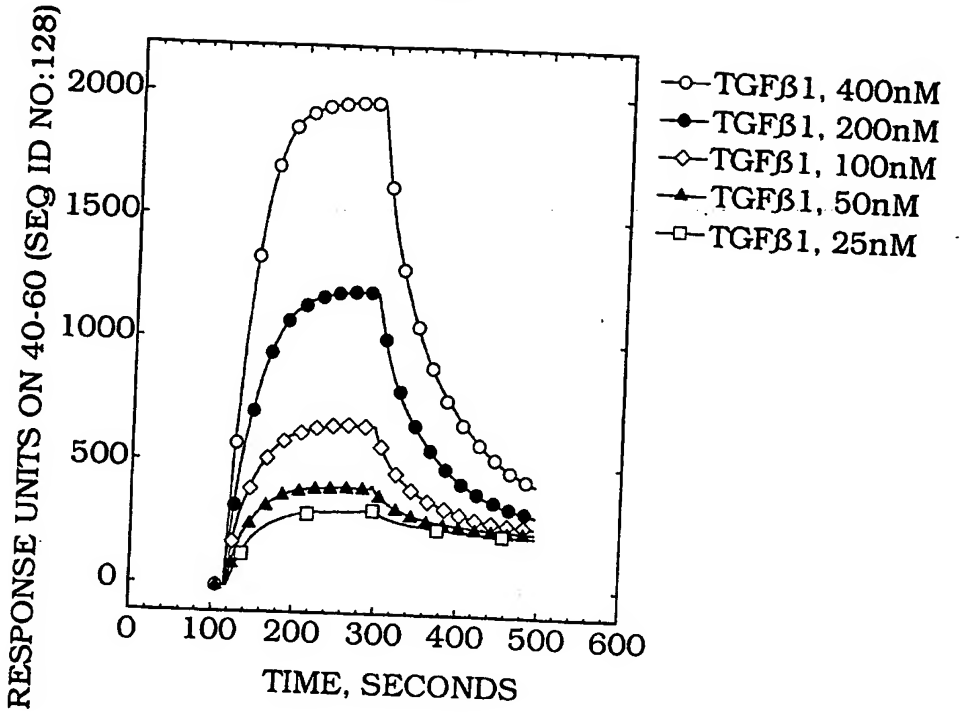

Figure 3C

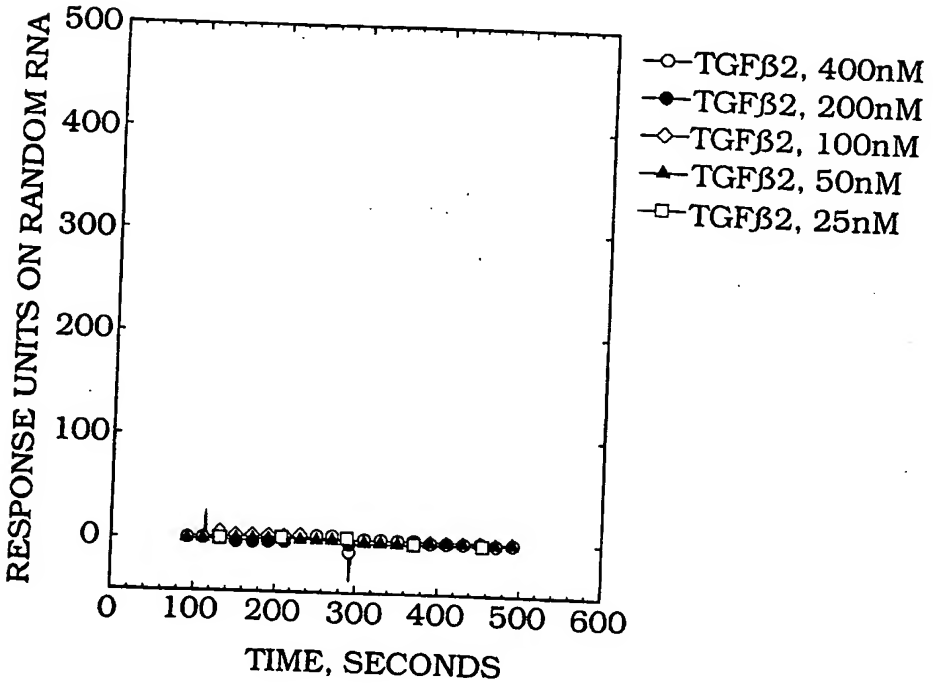

Figure 3D

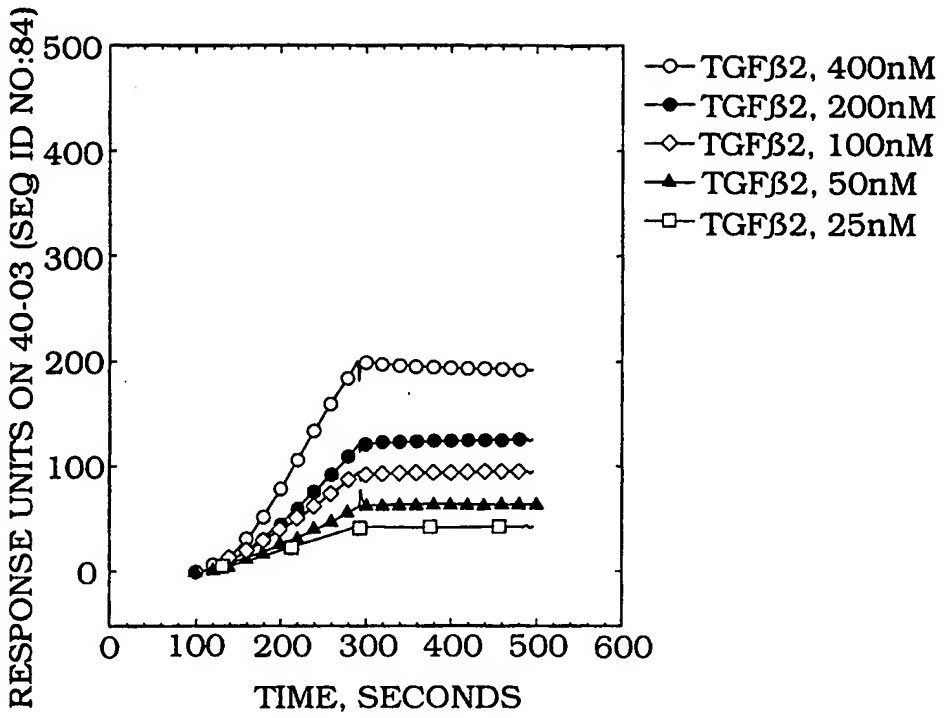

Figure 3E

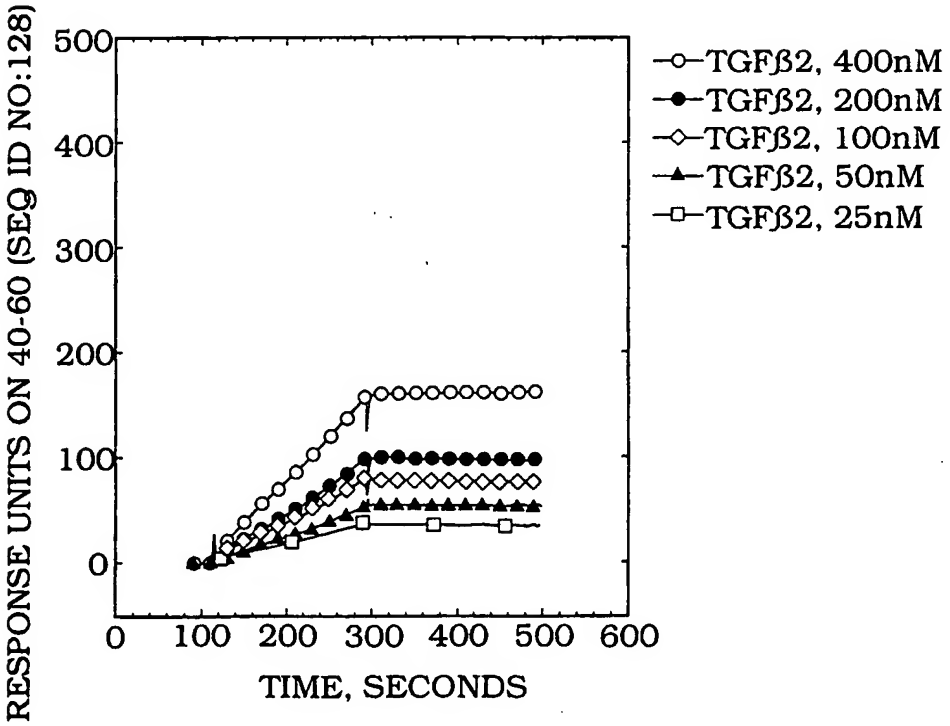

Figure 3F

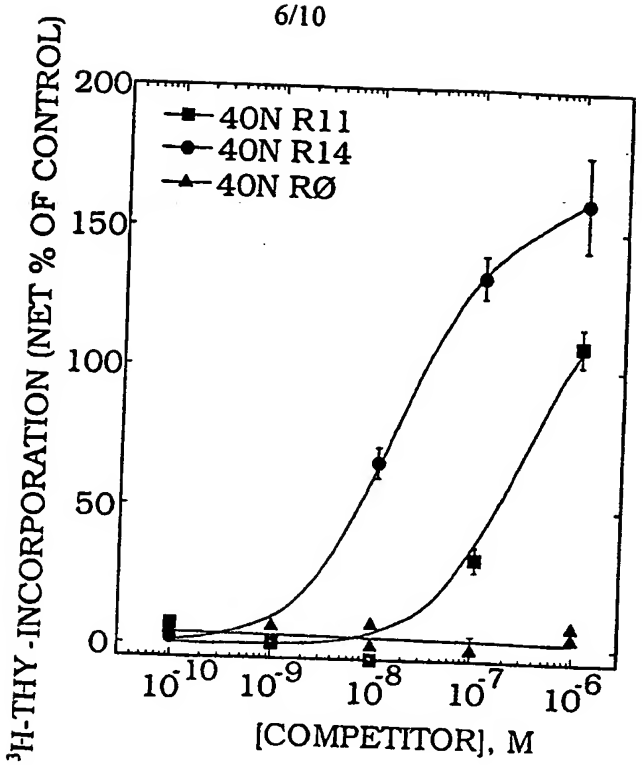

Figure 4A

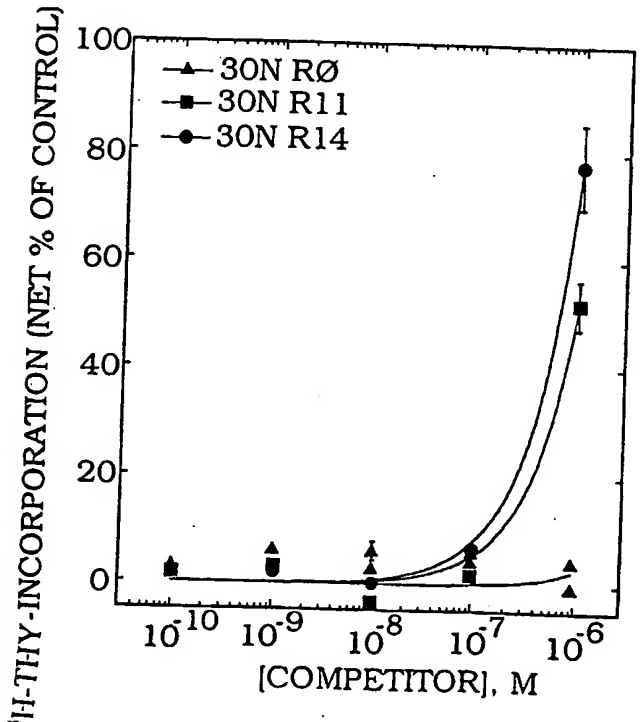

Figure 4B

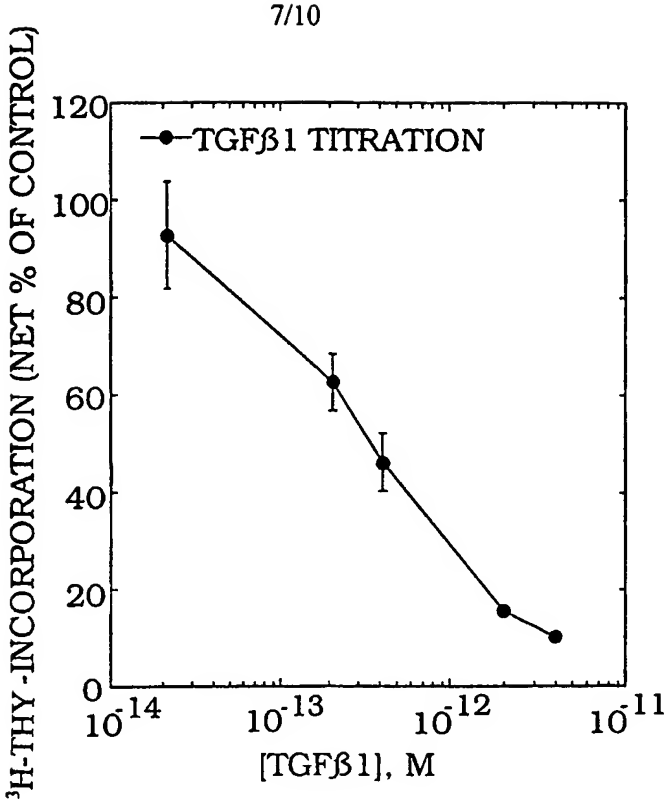

Figure 5A

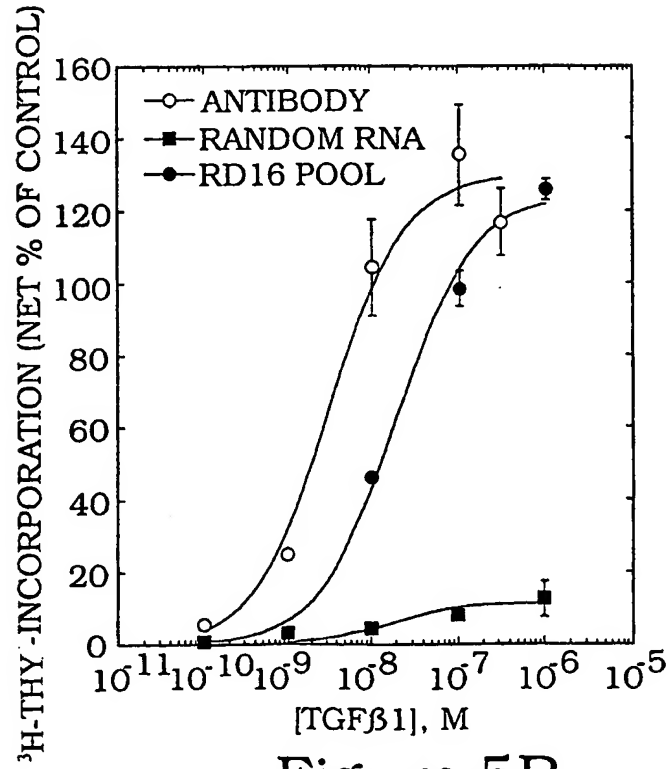

Figure 5B

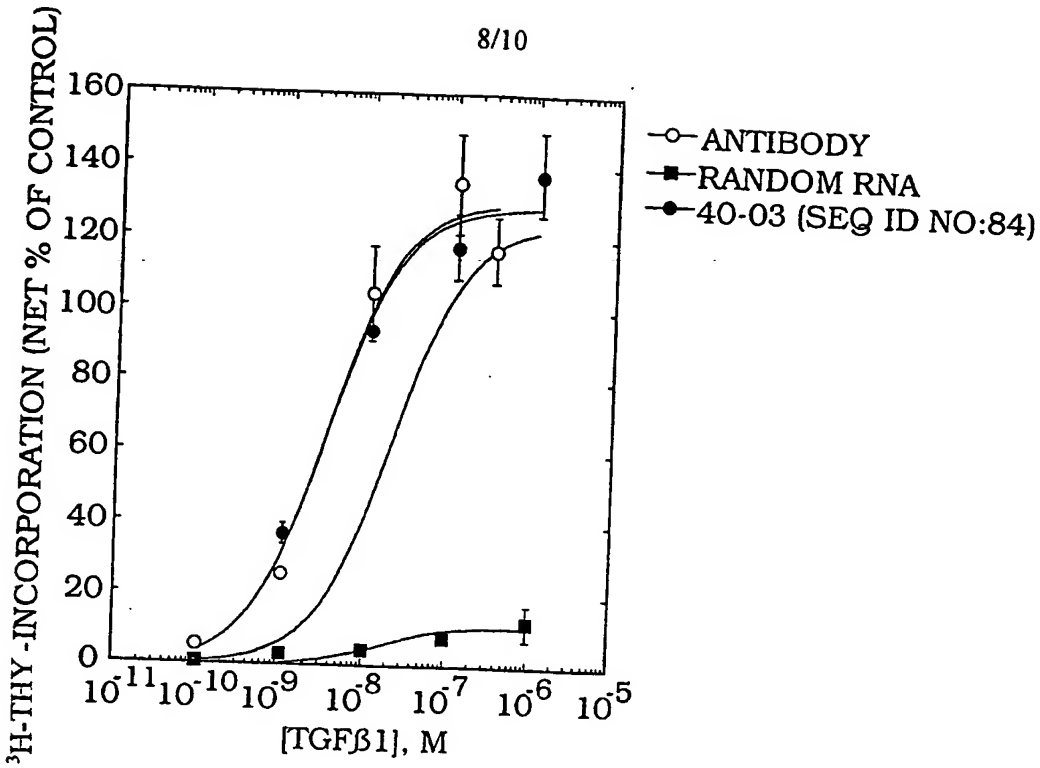

Figure 5C

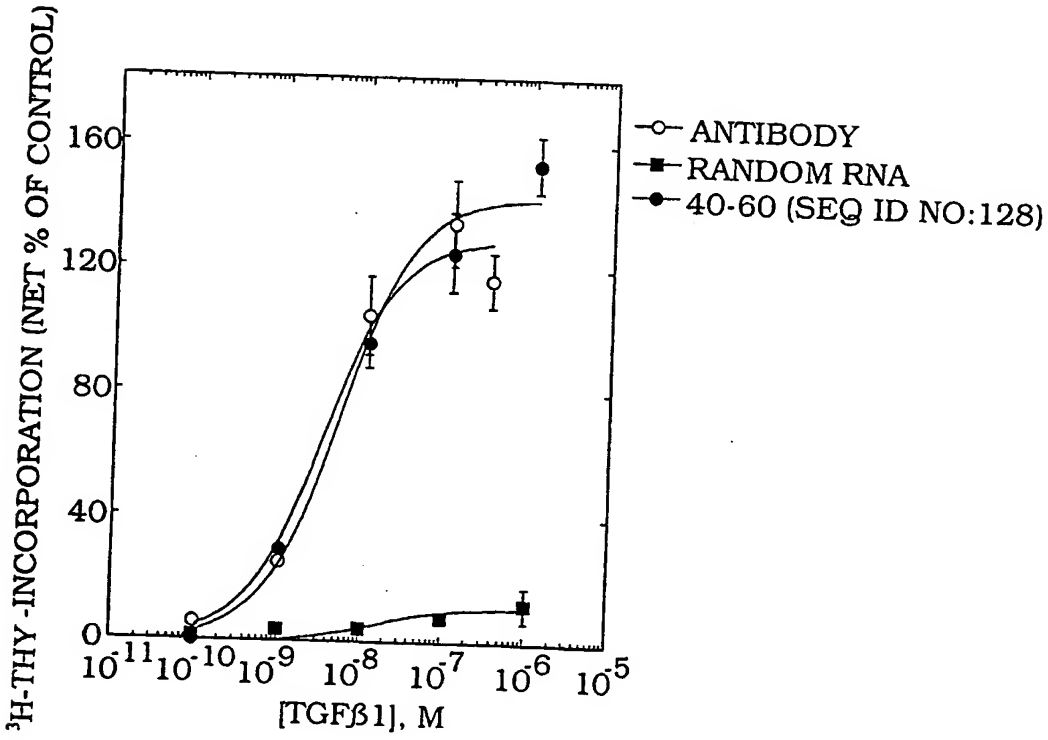

Figure 5D

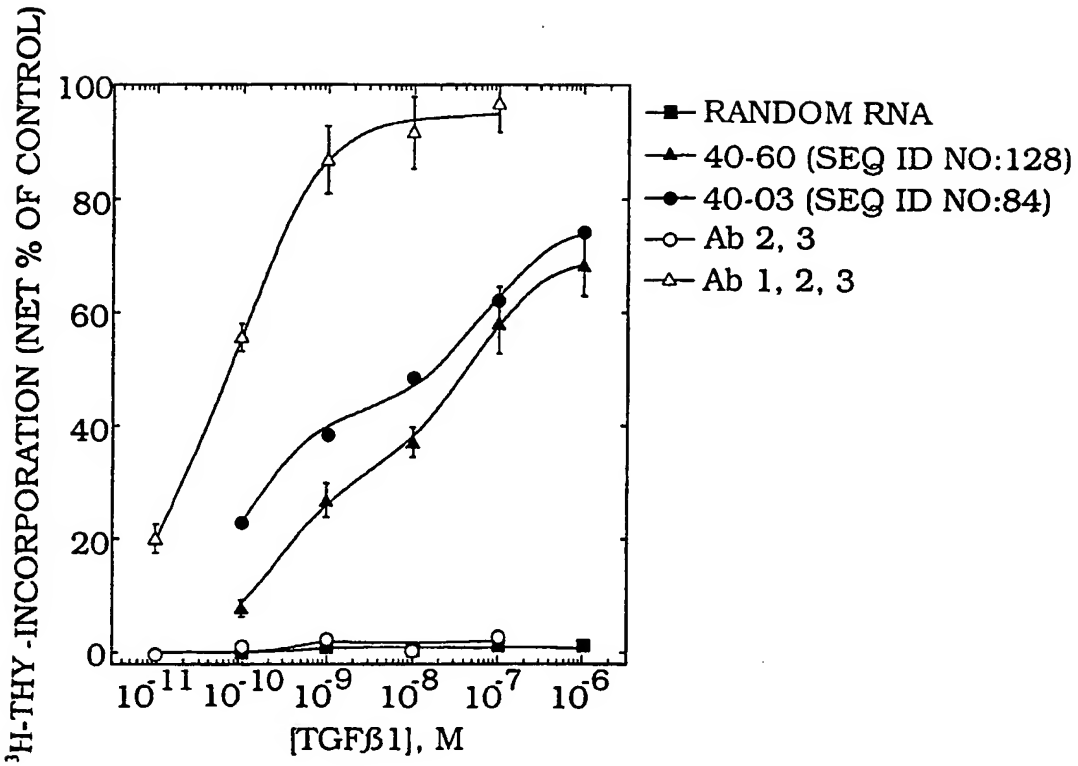
^{3}H-THY -INCORPORATION (NET % OF CONTROL)
100
80
60
40
20
0
10^{-11} 10^{-10} 10^{-9} 10^{-8} 10^{-7} 10^{-6}
[TGFβ1], M
RANDOM RNA
40-60 (SEQ ID NO:128)
40-03 (SEQ ID NO:84)
Ab 2, 3
Ab 1, 2, 3

Figure 6

```
                S2          S1          S3          S1          S2          S3
40-03   gggaggacgatgcggGGGTTA TTGGGCGTCAACATCCCCG ATTCTTTTCACGTCc  agacgactcgcccga
40-55      gggaggacgatgcggTTAG  GGGCGTCAACACCGCTATTCAACCTTCG NTTCCC cagacgactcgcccga
40-58      gggaggacgatgcggTTAT  GGGCGTCAACACCGCTATTACAACTTTCGCCTCCC cagacgactcgcccga
40-14 gggaggacgatg    cggCATTAT   GGCGTCAACATGCCGGTTTTCGATTCT CATTGTCcagacgactcgcccga
40-16      gggaggacgatgcggTTA   GGGGCGTCAACACCGCTATTACATCTTTCGCCTCCC cagacgac tcgcccga
40-19      gggaggacgatgcggTTAGCGCGAGTTCAACACCGCATGTGATTCTTTCGCCTCCc  agac    gactcgcccga
40-23      gggaggacgatgcggTTAG  GGGCGTCAACACCGCTATTACAATCTTCGCTTCCc  agacgactcgcccga
40-31      gggaggacgatgcggTTAA  GGGCGTCAACACCGCTATTACAACTTTCGCTTCCa  agacgactcgcccga
40-54      gggaggacgatgcggTTAG  GGGCGTCAACACCGCTATCATAACTTTCGCTTCCC cagacgactcgcccga
40-56      gggaggacgatgcggTTA   GGGCGTCAACACCGCTATTACAACTTTCGCNTCCC cagacgactcgcccga
40-60      gggaggacgatgcggTTAT  GGGCGTCAACACCGCTATTACAGTTTTCGCCTCCCCcagacgactcgcccga
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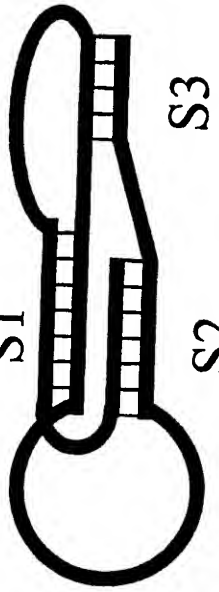

Fig. 7

SEQUENCE LISTING

(1) GENERAL INFORMATION:
(i) APPLICANT: LARRY GOLD
NIKOS PAGRATIS
(ii) TITLE OF THE INVENTION: HIGH AFFINITY TGFβ NUCLEIC ACID LIGANDS AND INHIBITORS
(iii) NUMBER OF SEQUENCES: 143
(iv) CORRESPONDENCE ADDRESS:
(A) ADDRESSEE: Swanson and Bratschun, L.L.C.
(B) STREET: 8400 East Prentice Avenue, Suite #200
(C) CITY: Denver
(D) STATE: Colorado
(E) COUNTRY: USA
(F) ZIP: 80111
(v) COMPUTER READABLE FORM:
(A) MEDIUM TYPE: Diskette, 3.5 inch, 1.44 Mb storage
(B) COMPUTER: IBM compatible
(C) OPERATING SYSTEM: MS DOS
(D) SOFTWARE: Word 97
(vi) CURRENT APPLICATION DATA:
(A) APPLICATION NUMBER: PCT/US99/_____
(B) FILING DATE:
(C) CLASSIFICATION:
(vi) PRIOR APPLICATION DATA:
(A) APPLICATION NUMBER: 09/046,247
(B) FILING DATE: 23-MARCH-1998
(C) CLASSIFICATION:
(vi) PRIOR APPLICATION DATA:
(A) APPLICATION NUMBER: 08/458,424
(B) FILING DATE: 2-JUNE-1995
(C) CLASSIFICATION:
(vii) PRIOR APPLICATION DATA:
(A) APPLICATION NUMBER: 07/714,131
(B) FILING DATE: 10-JUNE-1991
(vii) PRIOR APPLICATION DATA:
(A) APPLICATION NUMBER: 07/536,428
(B) FILING DATE: 11-JUNE-1990
(vii) PRIOR APPLICATION DATA:
(A) APPLICATION NUMBER: 07/964,624
(B) FILING DATE: 21-OCTOBER-1992
(vii) PRIOR APPLICATION DATA:
(A) APPLICATION NUMBER: 08/117,991
(B) FILING DATE: 8-SEPTEMBER-1993
(vii) PRIOR APPLICATION DATA:
(A) APPLICATION NUMBER: 07/931,473
(B) FILING DATE: 17-AUGUST-1992
(viii) ATTORNEY/AGENT INFORMATION:
(A) NAME: Barry Swanson
(B) REGISTRATION NUMBER: 33,215
(C) REFERENCE/DOCKET NUMBER: NEX 34.2/CIP-PCT
(ix) TELECOMMUNICATION INFORMATION:
(A) TELEPHONE: (303) 793-3333
(B) TELEFAX: (303) 793-3433
(2) INFORMATION FOR SEQUENCE ID NO: 1:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 71 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: DNA

(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 1:
GGGAGGACGA TGCGGNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN
NNNNNCAGAC GACTCGCCCG A 50
71

(2) INFORMATION FOR SEQUENCE ID NO: 2:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 61 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: DNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 2:
GGGAGGACGA TGCGGNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNCAGAC
GACTCGCCCG A 50
61

(2) INFORMATION FOR SEQUENCE ID NO: 3:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 51 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: DNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 3:
GGGAGGACGA TGCGGNNNNN NNNNNNNNNN NNNNNCAGAC GACTCGCCCG
A 50
51

(2) INFORMATION FOR SEQUENCE ID NO: 4:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 32 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: DNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 4:
TAATACGACT CACTATAGGG AGGACGATGC GG
32

(2) INFORMATION FOR SEQUENCE ID NO: 5:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 16 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: DNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 5:
TCGGGCGAGT CGTCTG
16

(2) INFORMATION FOR SEQUENCE ID NO: 6:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 51 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single

(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 6:

GGGAGGACGA UGCGGGUCUA UUUUUGCCUC CUCCCCAGAC GACUCGCCCG 50
A 51

(2) INFORMATION FOR SEQUENCE ID NO: 7:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 51 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 7:

GGGAGGACGA UGCGGAAUCC UUUCUUAAAC CUCCCCAGAC GACUCGCCCG 50
A 51

(2) INFORMATION FOR SEQUENCE ID NO: 8:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 61 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 8:

GGGAGGACGA UGCGGUGUCU UUAGCUUAGG UUAUUCCUUC UGCCGCAGAC 50
GACUCGCCCG A 61

(2) INFORMATION FOR SEQUENCE ID NO: 9:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 61 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 9:

GGGAGGACGA UGCGGUGUCU UUAGCUUAGG UGAUUCCUUC UGCCGCAGAC 50
GACUCGCCCG A 61

(2) INFORMATION FOR SEQUENCE ID NO: 10:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 62 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 10:

GGGAGGACGA UGCGGUGUCU CUACCUUAGG UUGAUUCCUU CUACCGCAGA 50
CGACUCGCCC GA 62

(2) INFORMATION FOR SEQUENCE ID NO: 11:

(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 50 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 11:
GGGAGGACGA UGCGGUGAGU CUUGUUUUUU CGUCCAGACG ACUCGCCCGA 50

(2) INFORMATION FOR SEQUENCE ID NO: 12:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 61 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 12:
GGGAGGACGA UGCGGUUGGC AUUGAAAGAG CUGGCAUACA UUCGCCAGAC 50
GACUCGCCCG A 61

(2) INFORMATION FOR SEQUENCE ID NO: 13:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 51 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 13:
GGGAGGACGA UGCGGUCCUU UCUAACAUUC CUCCCCAGAC GACUCGCCCG 50
A 51

(2) INFORMATION FOR SEQUENCE ID NO: 14:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 51 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 14:
GGGAGGACGA UGCGGGUCGU UGUUUUUCUC CUCCCCAGAC GACUCGCCCG 50
A 51

(2) INFORMATION FOR SEQUENCE ID NO: 15:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 51 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 15:

GGGAGGACGA UGCGGUGAGU CUUUCUUUUC GUCCCCAGAC GACUCGCCCG 50
A 51

(2) INFORMATION FOR SEQUENCE ID NO: 16:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 49 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 16:
GGGAGGACGA UGCGGGUCGU UUUUUUGGUC CUCCAGACGA CUCGCCCGA 49

(2) INFORMATION FOR SEQUENCE ID NO: 17:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 51 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 17:
GGGAGGACGA UGCGGGUUUU UAUUAUUCGU UUGGCCAGAC GACUCGCCCG 50
A 51

(2) INFORMATION FOR SEQUENCE ID NO: 18:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 52 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 18:
GGGAGGACGA UGCGGGUCGA UCAUUUUUAG CCUCCCCAGA CGACUCGCCC 50
GA 52

(2) INFORMATION FOR SEQUENCE ID NO: 19:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 51 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 19:
GGGAGGACGA UGCGGUGAGU UGAUCUUUUC GUCCCCAGAC GACUCGCCCG 50
A 51

(2) INFORMATION FOR SEQUENCE ID NO: 20:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 60 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA

(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 20:
GGGAGGACGA UGCGGUGCCU UUAGCUUAGG CAUUGCCUUC UGUGCAGACG 50
ACUCGCCCGA 60

(2) INFORMATION FOR SEQUENCE ID NO: 21:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 61 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 21:
GGGAGGACGA UGCGGCAAAA UUUUUGGUCA AGCCGUCAUU GCCGCCAGAC 50
GACUCGCCCG A 61

(2) INFORMATION FOR SEQUENCE ID NO: 22:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 51 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 22:
GGGAGGACGA UGCGGGUCGU UCUUUUUUCC CUCCCCAGAC GACUCGCCCG 50
A 51

(2) INFORMATION FOR SEQUENCE ID NO: 23:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 61 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 23:
GGGAGGACGA UGCGGAAUUU UUGUGAAGAC GUUUGCCGCU UUGCCCAGAC 50
GACUCGCCCG A 61

(2) INFORMATION FOR SEQUENCE ID NO: 24:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 51 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 24:
GGGAGGACGA UGCGGCGCAU CUUCUGUUUU CUCCCCAGAC GACUCGCCCG 50
A 51

(2) INFORMATION FOR SEQUENCE ID NO: 25:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 60 base pairs

(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 25:

GGGAGGACGA UGCGGGGAAU UUUUGGUAAA GCCGUAUGCC UCGCCAGACG 50
ACUCGCCCGA 60

(2) INFORMATION FOR SEQUENCE ID NO: 26:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 61 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 26:

GGGAGGACGA UGCGGUCAUC UCUGGGAGUU AAGAUCAUUU GGCCGCAGAC 50
GACUCGCCCG A 61

(2) INFORMATION FOR SEQUENCE ID NO: 27:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 51 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 27:

GGGAGGACGA UGCGGGCAGC CUCUGAUUUU CUCCCCAGAC GACUCGCCCG 50
A 51

(2) INFORMATION FOR SEQUENCE ID NO: 28:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 51 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 28:

GGGAGGACGA UGCGGGUCGU GAUUUUCGUU CUGCCCAGAC GACUCGCCCG 50
A 51

(2) INFORMATION FOR SEQUENCE ID NO: 29:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 52 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 29:

GGGAGGACGA UGCGGGUCGU AUUUUUUCCG CCUCCCCAGA CGACUCGCCC 50
GA 52

(2) INFORMATION FOR SEQUENCE ID NO: 30:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 59 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 30:
GGGAGGACGA UGCGGUCCUC AGCCUCUCAC UUAUUAUCCU CCCCAGACGA 50
CUCGCCCGA 59

(2) INFORMATION FOR SEQUENCE ID NO: 31:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 51 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 31:
GGGAGGACGA UGCGGGUCUA CUUGUUUUAC CUCCCCAGAC GACUCGCCCG 50
A 51

(2) INFORMATION FOR SEQUENCE ID NO: 32:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 51 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 32:
GGGAGGACGA UGCGGCGAUU UUUUCGUCUU UUGGCCAGAC GACUCGCCCG 50
A 51

(2) INFORMATION FOR SEQUENCE ID NO: 33:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 61 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 33:
GGGAGGACGA UGCGGUGUCU AUAGCCUUGA UUAUAUCAUC UGCCGCAGAC 50
GACUCGCCCG A 61

(2) INFORMATION FOR SEQUENCE ID NO: 34:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 51 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:

(D) OTHER INFORMATION: All pyrimidines are 2'-F modified

(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 34:

GGGAGGACGA UGCGGCGAUU CCUCUUUUCA CUCCCCAGAC GACUCGCCCG 50
A 51

(2) INFORMATION FOR SEQUENCE ID NO: 35:

(i) SEQUENCE CHARACTERIZATION:

(A) LENGTH: 51 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear

(ii) MOLECULAR TYPE: RNA

(ix) FEATURE:

(D) OTHER INFORMATION: All pyrimidines are 2'-F modified

(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 35:

GGGAGGACGA UGCGGUCCCA UUUUUCUCCU CUCCCCAGAC GACUCGCCCG 50
A 51

(2) INFORMATION FOR SEQUENCE ID NO: 36:

(i) SEQUENCE CHARACTERIZATION:

(A) LENGTH: 51 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear

(ii) MOLECULAR TYPE: RNA

(ix) FEATURE:

(D) OTHER INFORMATION: All pyrimidines are 2'-F modified

(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 36:

GGGAGGACGA UGCGGGUUAA UUUUUGUCCU CUGGCCAGAC GACUCGCCCG 50
A 51

(2) INFORMATION FOR SEQUENCE ID NO: 37:

(i) SEQUENCE CHARACTERIZATION:

(A) LENGTH: 56 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear

(ii) MOLECULAR TYPE: RNA

(ix) FEATURE:

(D) OTHER INFORMATION: All pyrimidines are 2'-F modified

(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 37:

GGGAGGACGA UGCGGUUUUU UUCUUUUUUC UUUUUUUCCG CAGACGACUC 50
GCCCGA 56

(2) INFORMATION FOR SEQUENCE ID NO: 38:

(i) SEQUENCE CHARACTERIZATION:

(A) LENGTH: 50 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear

(ii) MOLECULAR TYPE: RNA

(ix) FEATURE

(D) OTHER INFORMATION: All pyrimidines are 2'-F modified

(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 38:

GGGAGGACGA UGCGGUCGUC UUUGUUUUUC UCCCCAGACG ACUCGCCCGA 50

(2) INFORMATION FOR SEQUENCE ID NO: 39:

(i) SEQUENCE CHARACTERIZATION:

(A) LENGTH: 61 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single

(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 39:
GGGAGGACGA UGCGGUGUCU AUAGCCUUGA UUACAUCAUC UGCCGCAGAC 50
GACUCGCCCG A 61

(2) INFORMATION FOR SEQUENCE ID NO: 40:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 61 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 40:
GGGAGGACGA UGCGGUGCCU UUAGCUUAGG CAUUGCCUUC UGCCGCAGAC 50
GACUCGCCCG A 61

(2) INFORMATION FOR SEQUENCE ID NO: 41:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 61 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 41:
GGGAGGACGA UGCGGUGUCU AUAGCUUGAU UUUUAAUUUC UGCCGCAGAC 50
GACUCGCCCG A 61

(2) INFORMATION FOR SEQUENCE ID NO:42 :
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 51 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 42:
GGGAGGACGA UGCGGUUUUA UUUUCUUCGU CUGGCCAGAC GACUCGCCCG 50
A 51

(2) INFORMATION FOR SEQUENCE ID NO: 43:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 73 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 43:
GGGAGGACGA UGCGGGAUGA ACCGAACCGA GGUUAAGGUG CCAGAGUAGA 50
CGCUCAUCAG ACGACUCGCC CGA 73

(2) INFORMATION FOR SEQUENCE ID NO: 44:

(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 51 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 44:

GGGAGGACGA UGCGGUCGUC UAUUUUUUCC CUCCCCAGAC GACUCGCCCG 50
A 51

(2) INFORMATION FOR SEQUENCE ID NO: 45:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 51 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 45:

GGGAGGACGA UGCGGCUUUC GUCUGUUUUC CUGCCCAGAC GACUCGCCCG 50
A 51

(2) INFORMATION FOR SEQUENCE ID NO: 46:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 61 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 46:

GGGAGGACGA UGCGGUGUCU UUAGCCUAGG UGAUUCCUUC UGCCGCAGAC 50
GACUCGCCCG A 61

(2) INFORMATION FOR SEQUENCE ID NO: 47:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 60 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 47:

GGGAGGACGA UGCGGCCUUG UUUUCUUUUU UCUUUUUUCA CCCCCAGACG 50
ACUCGCCCGA 60

(2) INFORMATION FOR SEQUENCE ID NO: 48:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 61 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 48:

GGGAGGACGA UGCGGUGUCU UUAGCCCAGG UGAUUCCUUC UGCCGCAGAC 50
GACUCGCCCG A 61

(2) INFORMATION FOR SEQUENCE ID NO: 49:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 61 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 49:
GGGAGGACGA UGCGGUUAAC CGUAAAGACG GCAUGAUGUA GUCCGCAGAC 50
GACUCGCCCG A 61

(2) INFORMATION FOR SEQUENCE ID NO: 50:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 61 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 50:
GGGAGGACGA UGCGGUUUUU UUAGCUUAGG UGAUUCCUUC NNCCUCAGAC 50
GACUCGCCCG A 61

(2) INFORMATION FOR SEQUENCE ID NO: 51:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 61 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 51:
GGGAGGACGA UGCGGUGCCU UUAGCUUAGG CUUUGCCUUC UGCCGCAGAC 50
GACUCGCCCG A 61

(2) INFORMATION FOR SEQUENCE ID NO: 52:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 58 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 52:
GGGAGGACGA UGCGGCGGAA UUUUUGUUGA GCCGUAUGCC GCCAGACGAC 50
UCGCCCGA 58

(2) INFORMATION FOR SEQUENCE ID NO: 53:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 61 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear

(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 53:

GGGAGGACGA UGCGGUGCCU UUAGCUUAGG UGAUUCCUUC UGCCGCAGAC 50
GACUCGCCCG A 61

(2) INFORMATION FOR SEQUENCE ID NO: 54:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 61 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 54:

GGGAGGACGA UGCGGUGUCU UUAGCCUAGG UGAUUCCUUC UGCCGCAGAC 50
GACUCGCCCG A 61

(2) INFORMATION FOR SEQUENCE ID NO: 55:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 61 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 55:

GGGAGGACGA UGCGGUGUCU AUAGCCUGAU UUUUAAUCUC UGCCGCAGAC 50
GACUCGCCCG A 61

(2) INFORMATION FOR SEQUENCE ID NO: 56:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 61 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 56:

GGGAGGACGA UGCGGUUGAC CGUUAAGACG GCAUGAUGUG GUCCGCAGAC 50
GACUCGCCCG A 61

(2) INFORMATION FOR SEQUENCE ID NO: 57:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 61 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 57:

GGGAGGACGA UGCGGUGCCU UUAGCUUAGG CAUUGCCUUC UGCCGCAGAC 50
GACUCGCCCG A 61

(2) INFORMATION FOR SEQUENCE ID NO: 58:
(i) SEQUENCE CHARACTERIZATION:

(A) LENGTH: 61 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 58:
GGGAGGACGA UGCGGUGCCU UUAGCUUAGG CUUUGCCUUC UGCCGCAGAC 50
GACUCGCCCG A 61

(2) INFORMATION FOR SEQUENCE ID NO: 59:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 61 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 59:
GGGAGGACGA UGCGGUUAAC CNUAAAUACG GCUUGANUUC UUCCGCAGAC 50
GACUCGCCCG A 61

(2) INFORMATION FOR SEQUENCE ID NO: 60:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 61 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 60:
GGGAGGACGA UGCGGUGCCU UUAGCUUAGG CAUUGCCUUC UGCCGCAGAC 50
GACUCGCCCG A 61

(2) INFORMATION FOR SEQUENCE ID NO: 61:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 61 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 61:
GGGAGGACGA UGCGGUUAAC CGUAAAGACG GCAUGAUGUU UUCCGCAGAC 50
GACUCGCCCG A 61

(2) INFORMATION FOR SEQUENCE ID NO: 62:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 61 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 62:
GGGAGGACGA UGCGGUUGGC AUUGAAAGAG GCGUCAUAUG UUCGCCAGAC 50

GACUCGCCCG A 61

(2) INFORMATION FOR SEQUENCE ID NO: 63:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 62 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 63:
GGGAGGACGA UGCGGCCUUU CUUUCUUUUU AUUUUCUUCC CCUCCCCAGA 50
CGACUCGCCC GA 62

(2) INFORMATION FOR SEQUENCE ID NO: 64:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 61 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 64:
GGGAGGACGA UGCGGUGCCU UUAGCCUAGA CCUUGUCUUC UGCCGCAGAC 50
GACUCGCCCG A 61

(2) INFORMATION FOR SEQUENCE ID NO: 65:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 61 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 65:
GGGAGGACGA UGCGGUGUCU UUAGCCUAGG UGAUUCCUUC UGCCGCAGAC 50
GACUCGCCCG A 61

(2) INFORMATION FOR SEQUENCE ID NO: 66:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 61 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 66:
GGGAGGACGA UGCGGUGUCU UUAGCCUAGG UGAUUCCUUC UGCCGCAGAC 50
GACUCGCCCG A 61

(2) INFORMATION FOR SEQUENCE ID NO: 67:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 71 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA

(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 67:
GGGAGGACGA UGCGGACCGG UAAGGGCACU GCAGGAACAC AAUCCCCUAU 50
GCGACCAGAC GACUCGCCCG A 71

(2) INFORMATION FOR SEQUENCE ID NO:68 :
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 60 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 68:
GGGAGGACGA UGCGGGGAAU UUUUGGUAAA GCCGUAUGCC UCGCCAGACG 50
ACUCGCCCGA 60

(2) INFORMATION FOR SEQUENCE ID NO: 69:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 60 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 69:
GGGAGGACGA UGCGGUGGCA UUGAAAGAGA UCGCAUACCU UCGCCAGACG 50
ACUCGCCCGA 60

(2) INFORMATION FOR SEQUENCE ID NO: 70:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 61 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 70:
GGGAGGACGA UGCGGUGUCU AUAGCCUUGA UUACAUCAUC UGCCUCAGAC 50
GACUCGCCCG A 61

(2) INFORMATION FOR SEQUENCE ID NO: 71:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 61 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 71:
GGGAGGACGA UGCGGUGUCU UUAGCCUAGG UGAUUCCUUC UGCCUCAGAC 50
GACUCGCCCG A 61

(2) INFORMATION FOR SEQUENCE ID NO: 72:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 61 base pairs

(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 72:

GGGAGGACGA UGCGGUGCCU UUAGCUUAUG CAUUGCCUUC UGCCGCAGAC 50
GACUCGCCCG A 61

(2) INFORMATION FOR SEQUENCE ID NO: 73:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 62 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 73:

GGGAGGACGA UGCGGUGCCU UUAGCUUAGG CAUUCGCCUU CUGCCGCAGA 50
CGACUCGCCC GA 62

(2) INFORMATION FOR SEQUENCE ID NO: 74:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 61 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 74:

GGGAGGACGA UGCGGUGUCU UUGGCCUAGG UGAUUCCUUC UGCCGCAGAC 50
GACUCGCCCG A 61

(2) INFORMATION FOR SEQUENCE ID NO: 75:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 61 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 75:

GGGAGGACGA UGCGGUGUCU UUAGCUUAGG UGAUUCCUUC UGCCGCAGAC 50
GACUCGCCCG A 61

(2) INFORMATION FOR SEQUENCE ID NO: 76:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 61 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 76:

GGGAGGACGA UGCGGUGUCU UUAGCCUAGG UGAUUCCUUC UGCCGCAGAC 50
GACUCGCCCG A 61

(2) INFORMATION FOR SEQUENCE ID NO: 77:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 59 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 77:
GGGAGGACGA UGCGGUGCCU UUAGCUUAGG CAUUGCCUUG CCGCAGACGA 50
CUCGCCCGA 59

(2) INFORMATION FOR SEQUENCE ID NO: 78:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 62 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 78:
GGGAGGACGA UGCGGGGUCU UUUAUUUUUU GUUUUUCUCU GUGCCCCAGA 50
CGACUCGCCC GA 62

(2) INFORMATION FOR SEQUENCE ID NO: 79:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 61 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 79:
GGGAGGACGA UGCGGUUAAC CGUAAAGACA GCAUGAUGUA GUCUGCAGAC 50
GACUCGCCCG A 61

(2) INFORMATION FOR SEQUENCE ID NO: 80:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 60 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 80:
GGGAGGACGA UGCGGUUUUU UUCUUUUCCU UCCUUUUCUU ACCGCAGACG 50
ACUCGCCCGA 60

(2) INFORMATION FOR SEQUENCE ID NO: 81:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 61 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:

(D) OTHER INFORMATION: All pyrimidines are 2'-F modified

(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 81:

GGGAGGACGA UGCGGUUAAC CGUAAAGACG GCAUGAUGUU GUCCGCAGAC 50
GACUCGCCCG A 61

(2) INFORMATION FOR SEQUENCE ID NO: 82:

(i) SEQUENCE CHARACTERIZATION:

(A) LENGTH: 60 base pairs

(B) TYPE: nucleic acid

(C) STRANDEDNESS: single

(D) TOPOLOGY: linear

(ii) MOLECULAR TYPE: RNA

(ix) FEATURE:

(D) OTHER INFORMATION: All pyrimidines are 2'-F modified

(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 82:

GGGAGGACGA UGCGGGGAAU UUUUGGUAAA GCCGUAUGCC UCGCCAGACG 50
ACUCGCCCGA 60

(2) INFORMATION FOR SEQUENCE ID NO: 83:

(i) SEQUENCE CHARACTERIZATION:

(A) LENGTH: 72 base pairs

(B) TYPE: nucleic acid

(C) STRANDEDNESS: single

(D) TOPOLOGY: linear

(ii) MOLECULAR TYPE: RNA

(ix) FEATURE:

(D) OTHER INFORMATION: All pyrimidines are 2'-F modified

(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 83:

GGGAGGACGA UGCGGGCCAA GGUUACGCCG UCGGACCCUG CUGCCAACAU 50
CCUCCCCAGA CGACUCGCCC GA 72

(2) INFORMATION FOR SEQUENCE ID NO: 84:

(i) SEQUENCE CHARACTERIZATION:

(A) LENGTH: 70 base pairs

(B) TYPE: nucleic acid

(C) STRANDEDNESS: single

(D) TOPOLOGY: linear

(ii) MOLECULAR TYPE: RNA

(ix) FEATURE:

(D) OTHER INFORMATION: All pyrimidines are 2'-F modified

(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 84:

GGGAGGACGA UGCGGGGGUU AUUGGGCGUC AACAUCCCCG AUUCUUUUCA 50
CGUCCAGACG ACUCGCCCGA 70

(2) INFORMATION FOR SEQUENCE ID NO: 85:

(i) SEQUENCE CHARACTERIZATION:

(A) LENGTH: 71 base pairs

(B) TYPE: nucleic acid

(C) STRANDEDNESS: single

(D) TOPOLOGY: linear

(ii) MOLECULAR TYPE: RNA

(ix) FEATURE:

(D) OTHER INFORMATION: All pyrimidines are 2'-F modified

(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 85:

GGGAGGACGA UGCGGAUGCC UUUUGCCUUC AGGGUGUAAU UCCUUGAUCU 50
GUCCGCAGAC GACUCGCCCG A 71

(2) INFORMATION FOR SEQUENCE ID NO: 86:

(i) SEQUENCE CHARACTERIZATION:

(A) LENGTH: 72 base pairs

(B) TYPE: nucleic acid

(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 86:
GGGAGGACGA UGCGGAACAA GGUUACGCCG UCGGACCCUG CUGCCAACAU 50
CCUCCCCAGA CGACUCGCCC GA 72

(2) INFORMATION FOR SEQUENCE ID NO: 87:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 71 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 87:
GGGAGGACGA UGCGGUUAGG GGCGUCAACA CCGCUAUCAU AAUUUUCGCC 50
UUCCCCAGAC GACUCGCCCG A 71

(2) INFORMATION FOR SEQUENCE ID NO: 88:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 71 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 88:
GGGAGGACGA UGCGGCGCAA GGUUACGCCG UCGGACCCUG CUGCCAACAU 50
CCUCCCAGAC GACUCGCCCG A 71

(2) INFORMATION FOR SEQUENCE ID NO: 89:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 69 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 89:
GGGAGGACGA UGCGGUGCCU UUAGUCUGAA UCUUCUACCA UGAUUCUCUG 50
CCGCAGACGA CUCGCCCGA 69

(2) INFORMATION FOR SEQUENCE ID NO: 90:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 71 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 90:
GGGAGGACGA UGCGGGACCC UUGUCUGCGA UUCAACUCGU AGGUUUUCUC 50
ACGUGCAGAC GACUCGCCCG A 71

(2) INFORMATION FOR SEQUENCE ID NO: 91:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 72 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 91:

GGGAGGACGA UGCGGAGCAA GGUUACGAGG UCGGACCCUG CUGCCAACAU 50
CCUCCCCAGA CGACUCGCCC GA 72

(2) INFORMATION FOR SEQUENCE ID NO: 92:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 71 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 92:

GGGAGGACGA UGCGGCAUUA UGGCGUCAAC AUGCCGGUUU UCGAUUCUCA 50
UUGUCCAGAC GACUCGCCCG A 71

(2) INFORMATION FOR SEQUENCE ID NO: 93:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 70 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 93:

GGGAGGACGA UGCGGCUCUA ACUUCUUUUU CGCCUGUGUG UUUUCUUUUU 50
GCUGCAGACG ACUCGCCCGA 70

(2) INFORMATION FOR SEQUENCE ID NO: 94:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 70 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 94:

GGGAGGACGA UGCGGUUAGG GGCGUCAACA CCGCUAUUAC AUCUUUCGCC 50
UCCCCAGACG ACUCGCCCGA 70

(2) INFORMATION FOR SEQUENCE ID NO: 95:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 69 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified

(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 95:
GGGAGGACGA UGCGGGGUCG UUUUGUUUUU GUUUUUUGUA GCCCGGUCAU 50
CCCCAGACGA CUCGCCCGA 69

(2) INFORMATION FOR SEQUENCE ID NO: 96:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 71 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 96:
GGGAGGACGA UGCGGUUAGC GCGAGUUCAA CACCGCAUGU GAUUCUUUCG 50
CCUCCCAGAC GACUCGCCCG A 71

(2) INFORMATION FOR SEQUENCE ID NO: 97:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 72 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 97:
GGGAGGACGA UGCGGUACAA GGUUACGCCG UCGGACCCUG CUGCCAACAU 50
CCUCCCCAGA CGACUCGCCC GA 72

(2) INFORMATION FOR SEQUENCE ID NO: 98:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 70 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 98:
GGGAGGACGA UGCGGGACCC UUGUCUGCGA UUCAACUCGU AGGUCUUCUC 50
CGUGCAGACG ACUCGCCCGA 70

(2) INFORMATION FOR SEQUENCE ID NO: 99:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 69 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 99:
GGGAGGACGA UGCGGUUAGG GGCGUCAACA CCGCUAUUAC AAUUUUCGCU 50
UCCCAGACGA CUCGCCCGA 69

(2) INFORMATION FOR SEQUENCE ID NO: 100:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 69 base pairs
(B) TYPE: nucleic acid

(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 100:

GGGAGGACGA UGCGGUUAGG GGCGUCAACA CCGCUAUUAC AAUCUUCGCU 50
UCCCAGACGA CUCGCCCGA 69

(2) INFORMATION FOR SEQUENCE ID NO: 101:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 70 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 101:

GGGAGGACGA UGCGGUUAUG GGCGUCAACA CCGCUAUUAC AACUUUCGCU 50
UUCCCAGACG ACUCGCCCGA 70

(2) INFORMATION FOR SEQUENCE ID NO: 102:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 72 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 102:

GGGAGGACGA UGCGGUGUCG AUCGUUUGCU GUUUGAUUUC UUUUGUCCCU 50
CCCGUGCAGA CGACUCGCCC GA 72

(2) INFORMATION FOR SEQUENCE ID NO: 103:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 70 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 103:

GGGAGGACGA UGCGGUUAGG GGCGUCAACA UCGCUAUUAC AAUCUUCGCC 50
UUCCCAGACG ACUCGCCCGA 70

(2) INFORMATION FOR SEQUENCE ID NO: 104:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 70 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 104:

GGGAGGACGA UGCGGUUAGG GGCGUCAACA CCGCUAUUAC AACUUUCGCC 50
UCACCAGACG ACUCGCCCGA 70

(2) INFORMATION FOR SEQUENCE ID NO: 105:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 71 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 105:
GGGAGGACGA UGCGGGACCC UUUUCUGCGA UUCAACUCGU ACGUCUUCUC 50
ACGUGCAGAC GACUCGCCCG A 71

(2) INFORMATION FOR SEQUENCE ID NO: 106:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 68 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 106:
GGGAGGACGA UGCGGUUAAG GGCGUCAACA CCGCUAUUAA ACUUUCGCUU 50
CCCAGACGAC UCGCCCGA 68

(2) INFORMATION FOR SEQUENCE ID NO: 107:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 68 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 107:
GGGAGGACGA UGCGGUUAUG GGCGUCAACA CCGCUAUUAC AACUUUCGCC 50
UCCAGACGAC UCGCCCGA 68

(2) INFORMATION FOR SEQUENCE ID NO: 108:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 72 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 108:
GGGAGGACGA UGCGGAGCAA GGUUACGCCG UCGGACCCUG CUGCCAACAU 50
CCUCCCCAGA CGACUCGCCC GA 72

(2) INFORMATION FOR SEQUENCE ID NO: 109:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 63 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified

(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 109:
GGGAGGACGA UGCGGGUCAA GGUUACGCCG UCGGACCCUA CUGCCCCCAG 50
ACGACUCGCC CGA 63

(2) INFORMATION FOR SEQUENCE ID NO: 110:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 72 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 110:
GGGAGGACGA UGCGGCUCCU AUAUUCAUGU UAUUGUUUUU UUCUUCCAGC 50
UUGCCCCAGA CGACUCGCCC GA 72

(2) INFORMATION FOR SEQUENCE ID NO: 111:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 71 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 111:
GGGAGGACGA UGCGGAGAUA AUUAUCAGCG GUGGACGGGG UGCCGGUACU 50
GCCGCCAGAC GACUCGCCCG A 71

(2) INFORMATION FOR SEQUENCE ID NO: 112:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 69 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 112:
GGGAGGACGA UGCGGUGCCU UUAGCCUAAG UUGAUCUAUU CAGCUUUCUG 50
CCGCAGACGA CUCGCCCGA 69

(2) INFORMATION FOR SEQUENCE ID NO: 113:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 72 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified.
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 113:
GGGAGGACGA UGCGGCCCAA GGUUACGCCG UCGGACCCUA CUGCCAACUU 50
CCUCCCCAGA CGACUCGCCC GA 72

(2) INFORMATION FOR SEQUENCE ID NO: 114:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 70 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single

(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 114:
GGGAGGACGA UGCGGUGCCU UUAGCCUGAG UAUACUGAUG UAUAUUCUCU 50
GCCGCAGACG ACUCGCCCGA 70

(2) INFORMATION FOR SEQUENCE ID NO: 115:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 70 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 115:
GGGAGGACGA UGCGGUAGCG CGAGUUCAAC ACCGCAUGUG ACUCUUUCGC 50
CUCCCAGACG ACUCGCCCGA 70

(2) INFORMATION FOR SEQUENCE ID NO: 116:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 72 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 116:
GGGAGGACGA UGCGGAUCCU UUUUUUAGCU UUUUUCUUUU UCCUGCCCCA 50
CUUCCCCAGA CGACUCGCCC GA 72

(2) INFORMATION FOR SEQUENCE ID NO: 117:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 72 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 117:
GGGAGGACGA UGCGGUGCAA GGUUACGCCG UCGGACCCUG CUGCCAACAU 50
CCUCCCCAGA CGACUCGCCC GA 72

(2) INFORMATION FOR SEQUENCE ID NO: 118:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 69 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 118:
GGGAGGACGA UGCGGGGGCU UUUCCUUUAG UACUUUUUUG UUUCGCUCCC 50
CCCCAGACGA CUCGCCCGA 69

(2) INFORMATION FOR SEQUENCE ID NO: 119:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 69 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 119:

GGGAGGACGA UGCGGUGCCU UUAGUCUGAA UCUUACCAUG CGAUUUUCUG 50
CCGCAGACGA CUCGCCCGA 69

(2) INFORMATION FOR SEQUENCE ID NO: 120:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 72 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 120:

GGGAGGACGA UGCGGAACAA GGUUACUCCG UCGGACCCUG CUGCCAACAU 50
CCUCCCCAGA CGACUCGCCC GA 72

(2) INFORMATION FOR SEQUENCE ID NO: 121:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 71 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 121:

GGGAGGACGA UGCGGGACUC UUGUCUGCGA UUCAACUCGU AGGUCUUCUC 50
ACGUGCAGAC GACUCGCCCG A 71

(2) INFORMATION FOR SEQUENCE ID NO: 122:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 70 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 122:

GGGAGGACGA UGCGGUUAGG GGCGUCAACA CCGCUAUCAU AACUUUCGCU 50
UCCCCAGACG ACUCGCCCGA 70

(2) INFORMATION FOR SEQUENCE ID NO: 123:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 69 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified

(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 123:
GGGAGGACGA UGCGGUUAGG GGCGUCAACA CCGCUAUUCA ACCUUCGCUU 50
CCCCAGACGA CUCGCCCGA 69

(2) INFORMATION FOR SEQUENCE ID NO: 124:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 69 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 124:
GGGAGGACGA UGCGGUUAGG GCGUCAACAC CGCUAUUACA ACUUUCGCCU 50
CCCCAGACGAC UCGCCCGA 69

(2) INFORMATION FOR SEQUENCE ID NO: 125:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 51 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 125:
GGGAGGACGA UGCGGGGUGU CGUCUUUCAA CCCCUCAGAC GACUCGCCCG 50
A 51

(2) INFORMATION FOR SEQUENCE ID NO: 126:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 70 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 126:
GGGAGGACGA UGCGGUUAUG GGCGUCAACA CCGCUAUUAC AACUUUCGCC 50
UCCCCAGACG ACUCGCCCGA 70

(2) INFORMATION FOR SEQUENCE ID NO: 127:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 72 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 127:
GGGAGGACGA UGCGGCCCAA GGUUACGCCG UCGGACCCUG CUGCAAACAU 50
CCUCCCCAGA CGACUCGCCC GA 72

(2) INFORMATION FOR SEQUENCE ID NO: 128:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 71 base pairs
(B) TYPE: nucleic acid

(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 128:

GGGAGGACGA UGCGGUUAUG GGCGUCAACA CCGCUAUUAC AGUUUUCGCC 50
UCCCCCAGAC GACUCGCCCG A 71

(2) INFORMATION FOR SEQUENCE ID NO: 129:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 70 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 129:

GGGAGGACGA UGCGGUUAGG GGCGUCAACA CCGCUAUUAC AAUCUUCGCU 50
UUCCCAGACG ACUCGCCCGA 70

(2) INFORMATION FOR SEQUENCE ID NO: 130:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 72 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 130:

GGGAGGACGA UGCGGGCCAA GGUUACGCCG UCGGACCCUG CUGCCAACAU 50
CUUCCCCAGA CGACUCGCCC GA 72

(2) INFORMATION FOR SEQUENCE ID NO: 131:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 70 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 131:

GGGAGGACGA UGCGGUUAGG GGCGUCAACA CCGCUAUUAC AAUCUUCGUC 50
UUCCCAGACG ACUCGCCCGA 70

(2) INFORMATION FOR SEQUENCE ID NO: 132:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 72 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 132:

GGGAGGACGA UGCGGGUCAA GUUUACGCCG UCGGACCCUG CUGCCAACAU 50
CCUCCCCAGA CGACUCGCCC GA 72

(2) INFORMATION FOR SEQUENCE ID NO: 133:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 72 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 133:
GGGAGGACGA UGCGGUUCAA GGUUACGCCG UCGGACCCUG CUGCCAACAU 50
CCUCCCCAGA CGACUCGCCC GA 72

(2) INFORMATION FOR SEQUENCE ID NO: 134:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 72 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 134:
GGGAGGACGA UGCGGCUCAA GGUUACGCCG UCGGACCCUG CUGCCAACAU 50
CCUCCCCAGA CGACUCGCCC GA 72

(2) INFORMATION FOR SEQUENCE ID NO: 135:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 70 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 135:
GGGAGGACGA UGCGGUUAGG GGCUUCAACA CCGCUAUUAC AUUCUUCGCC 50
UCCCCAGACG ACUCGCCCGA 70

(2) INFORMATION FOR SEQUENCE ID NO: 136:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 72 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 136:
GGGAGGACGA UGCGGCACAA AGUUACGCCG UAGGACCCUG CUGCCAACAU 50
CCUCCCCAGA CGACUCGCCC GA 72

(2) INFORMATION FOR SEQUENCE ID NO: 137:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 72 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified

(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 137:
GGGAGGACGA UGCGGGGAUG GUCAGUUUCG GUUUUUCAUA UGUUUAUUUU 50
CCCCCCCAGA CGACUCGCCC GA 72

(2) INFORMATION FOR SEQUENCE ID NO: 138:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 66 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 138:
GGGAGGACGA UGCGGUAUUG ACUUUUGUUU CUUUUUCUUU GCCUGGUCCC 50
CAGACGACUC GCCCGA 66

(2) INFORMATION FOR SEQUENCE ID NO: 139:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 70 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 139:
GGGAGGACGA UGCGGUUAGG GGCGUCAACA CCGCUAUUAC AACUUUCGCU 50
UCCCCAGACG ACUCGCCCGA 70

(2) INFORMATION FOR SEQUENCE ID NO: 140:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 69 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 140:
GGGAGGACGA UGCGGCUUCU UUUUCUUCUU UUCUUUAUGU CUUCUUCAUG 50
CCGCAGACGA CUCGCCCGA 69

(2) INFORMATION FOR SEQUENCE ID NO: 141:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 71 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 141:
GGGAGGACGA UGCGGGACCN UUGUNUGCGA UUCAACUCGU AGGUCUUCUC 50
ACGUGCAGAC GACUCGCCCG A 71

(2) INFORMATION FOR SEQUENCE ID NO: 142:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 69 base pairs
(B) TYPE: nucleic acid

(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 142:

GGGAGGACGA UGCGGUUAUG GGCGUCAACA CCGCUAUUAC AACUUUCGCC 50
CCCCAGACGA CUCGCCCGA 69

(2) INFORMATION FOR SEQUENCE ID NO: 143:
(i) SEQUENCE CHARACTERIZATION:
(A) LENGTH: 70 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULAR TYPE: RNA
(ix) FEATURE:
(D) OTHER INFORMATION: All pyrimidines are 2'-F modified
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 143:

GGGAGGACGA UGCGGUUAUG GGUGUCAACA CCGCUAUUAC AACUUUCGCC 50
UCCCCAGACG ACUCGCCCGA 70

INTERNATIONAL SEARCH REPORT

International application No.
PCT/US99/05964

A. CLASSIFICATION OF SUBJECT MATTER

IPC(6) : C07H 21/02
US CL :536/24.3, 22.1, 23.1
According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)

. U.S. : 536/24.3, 22.1, 23.1

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)

GenEmbl, EST,N_Geneseq, Pending Patents_NA, Issued Patents_NA; search: OLIGO_NUC

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A,P	US 5,731,144 A (TOOTHMAN et al.) 24 March 1998, see entire document	1
A,P	US 5,731,424 A (TOOTHMAN et al.) 24 March 1998, see entire document, especially claims	1

☐ Further documents are listed in the continuation of Box C. ☐ See patent family annex.

*	Special categories of cited documents:	"T"	later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention
"A"	document defining the general state of the art which is not considered to be of particular relevance	"X"	document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone
"E"	earlier document published on or after the international filing date	"Y"	document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art
"L"	document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)	"&"	document member of the same patent family
"O"	document referring to an oral disclosure, use, exhibition or other means		
"P"	document published prior to the international filing date but later than the priority date claimed		

Date of the actual completion of the international search 08 MAY 1999	Date of mailing of the international search report 27 MAY 1999
Name and mailing address of the ISA/US Commissioner of Patents and Trademarks Box PCT Washington, D.C. 20231 Facsimile No. (703) 305-3230	Authorized officer STEPHANIE ZITOMER, PHD Telephone No. (703) 308-0196

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